Ag 3

United States
Environmental Protection
Agency

Office of
Radiation Programs
Washington, D.C. 20460

EPA 520/1-85-016
February 1986

Radiation

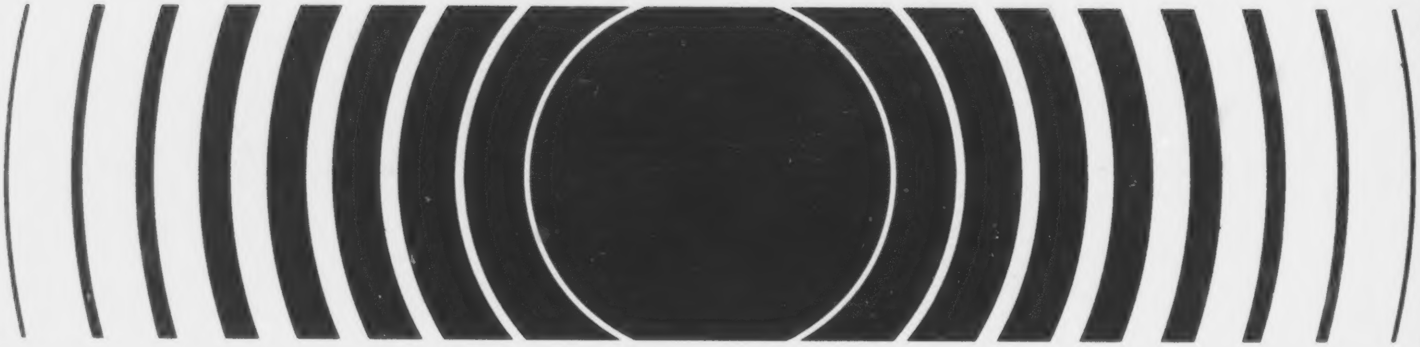
EPA

Effects of Radiation on Aquatic Organisms and Radiobiological Methodologies for Effects Assessment

NEWARK PUBLIC LIBRARY

MAY 20 1986

U. S. GOVT. PUBS.
DEPOSITORY 376



EP 6.2:

EPA520/1-85-016

Effects of Radiation on Aquatic Organisms and Radiobiological Methodologies for Effects Assessment

S.L. Anderson
F.L. Harrison

Work Performed Under
IAG DW 89930414-01

February 1986

U.S. Environmental Protection Agency
Office of Radiation Programs
Washington, D.C. 20460

FOREWORD

In response to the mandate of Public Law 92-532, the Marine Protection, Research and Sanctuaries Act of 1972, as amended, the Environmental Protection Agency (EPA) has developed a program to promulgate regulations and criteria to control the ocean disposal of radioactive wastes. An important technical consideration in any environmental assessment of this option is the potential for biological effects to marine organisms.

This report, prepared by Lawrence Livermore National Laboratory, summarizes the literature on effects to aquatic organisms from acute and chronic exposure to ionizing radiation for mortality, pathophysiology, reproduction, development, and genetic effects. Methodologies for the study of radiobiological effects are discussed, and recommendations for future research are provided. As such, these recommendations reflect only the views and opinions of the authors and have not been adopted by the Agency as necessary to meet any ocean disposal regulatory objectives.

The Office of Radiation Programs (ORP) will use this report to determine the direction of any future effects testing program. The methodologies described in the report may also be useful to those studying pollutants other than radiation.

The Agency invites all readers of this report to send any comments or suggestions to Mr. David E. Janes, Director, Analysis and Support Division, Office of Radiation Programs (ANR-461), Environmental Protection Agency, Washington, D.C. 20460.

Sheldon Meyers

Sheldon Meyers, Acting Director
Office of Radiation Programs (ANR-458)

ABSTRACT

This report reviews the results of studies on biological effects of ionizing radiation on aquatic organisms on the bases of biological effects (mortality, pathophysiology, reproduction, development, and genetics) and radiation regime (acute and chronic). The extensive data base is summarized in tables, but only the studies demonstrating effects from exposure to low levels of radiation are discussed critically because mortality as an endpoint is unsuitable for assessing the sublethal effects that may be expected from ocean disposal of low-level radioactive waste.

Results of studies of the effect of acute radiation on mortality indicate that the range of lethal levels in adult fish is 375 to 55,000 rad and that mortality of fish embryos has been demonstrated as low as 16 R. For invertebrates, lethal doses range from 210 rad to above 50,000 rad. For both fish and invertebrates, LD_{50} observation times are not standardized, and results may be modifed by other factors such as lifestage and temperature.

Research using pathophysiological endpoints on fish and invertebrates has mainly served to characterize lethal radiation syndromes. Further research in this area should focus on immune responses.

One of the few areas in aquatic radiobiology for which there are data on the effects of chronic, low-dose irradiation regimes is the study of reproductive effects in fish. Effects have been shown from 0.59 rad/d to 12 rad/d. These data indicate that the cellular sensitivity of some fishes is not less than that in mammals.

Developmental defects in embryos and embryo death induced by exposure below 100 R have been demonstrated for three species of fish. The lowest effect-level was 16 R. Much of the existing data on chronic irradiation of fish embryos is not useful because errors exist in the dosimetry for studies using radionuclides in the test water. However, an increase in opercular defects in Oncorhynchus tshawytscha was found at 0.5 R/d. Little information exists on developmental effects in invertebrates. However, decreased hatching rate of the snail Physa hyterostropha was observed at 10 rad/h.

Genotoxic effects in fish and invertebrates have been observed at low doses. Increased frequency of chromosomal aberrations in cultured fish cells has been observed after 50 R exposures. In the marine worm *Neanthes arenaceodentata*, significant increases in sister-chromatid exchange and frequency of chromosomal aberrations have been observed at 60 rad and 200 rad. An increased frequency of major eye malformations in trout embryos occurred after sperm were exposed to 25 rad. The data on genotoxic effects further demonstrate that the cellular-level sensitivity of certain aquatic organisms may not be significantly different from that of mammals.

Because the information on effects of low levels of radiation on aquatic organisms is limited, information on terrestrial animals is also discussed to provide greater perspective on potential effects at the dose rates in question. In addition, the limited information on effects on populations and communities from chronic irradiation is reviewed.

We conclude that no data are available demonstrating that significant detrimental effects on aquatic organisms occur at the maximal levels of contamination (0.240 rad/d at the end of the pipeline at Windscale) reported in the ocean due to waste-disposal activity. However, there are not sufficient data on aquatic organisms in the literature to determine threshold dose rates at which important radiobiological effects would be observed. Furthermore, the certainty of any prediction would be limited by our incomplete understanding of the effects of potentially modifying factors, especially temperature, on the responses of the organisms to low dose rates or total doses.

Important gaps in knowledge needed by regulators on the biological effects of ionizing radiation on aquatic organisms were identified, as follows: (1) few reliable studies at chronic and low dose rates have been conducted, (2) few studies have been done on marine organisms, (3) very few studies have been done on marine invertebrates, (4) information on modifying factors such as temperature, species specificity, and cell kinetics is very scarce, and (5) the long-term effects of low-level radiation on fertility in fish and invertebrates have not been adequately characterized.

General recommendations for future research include additional studies on marine invertebrates and reproductive and genotoxic endpoints in marine fishes and invertebrates. Further studies are also needed on the effects of modifying environmental factors, such as temperature. The additional methodologies for studying radiobiological effects include quantification of DNA single-strand breaks, unscheduled DNA synthesis, and premature chromosome condensation. Possible monitoring approaches that were given include using biological dosimetry such as quantification of chromosomal aberrations in a tissue that integrates effects and using the radiation-induced electron paramagnetic resonance signals in calcified tissues.

READEX FICHE

best copy available

CONTENTS

Abstract iii
List of Tables vii
List of Figures ix
Introduction 1
Stages in the Development and Modification of Radiation Injury 3
  Development of Radiation Injury 4
  Modification of Radiation Effects 6
  Principles of Human Radiation Protection and Their Relation to Aquatic Toxicology 9
Effects on Aquatic Animals of Acute and Chronic Exposure to Ionizing Radiation 11
  Mortality 11
    Criteria for Establishing a Lethal Dose 11
    Mortality of Fish from Acute Radiation Exposure 12
    Mortality of Invertebrates from Acute Radiation Exposure 20
    Mortality of Fish and Invertebrates from Chronic Radiation Exposure 24
    Conclusions on Mortality Data 24
  Pathophysiology 25
    Pathophysiological Effects in Fish from Acute Radiation Exposure 25
    Pathophysiological Effects in Invertebrates from Acute Radiation Exposure 32
    Pathophysiological Effects in Fish and Invertebrates from Chronic Radiation Exposure 32
    Conclusions on Pathophysiology Data 33
  Reproduction 35
    Reproductive Effects in Fish from Acute Radiation Exposure 35
    Reproductive Effects in Invertebrates from Acute Radiation Exposure 37
    Reproductive Effects in Fish and Invertebrates from Chronic Radiation Exposure 39
    Conclusions on Reproduction Data 43

Development 43

Developmental Effects from Acute and Chronic Irradiation of Fish 44

Developmental Effects from Acute and Chronic Irradiation of Invertebrates 50

Conclusions on Development Data 52

Genetics 53

General Considerations in Aquatic Genotoxicity Research 54

Induction of Chromosomal Aberrations in Fish 57

Induction of Mutations in Fish 60

Induction of Chromosomal Aberrations in Invertebrates 63

Induction of Mutations in Invertebrates 65

Conclusions on Genetics Data 66

Comparison of Radiation Effects on Aquatic Organisms to Those on Terrestrial Organisms 67

Mortality and Pathophysiology 67

Reproduction 69

Development 69

Genetics 69

Low-Level Radiation Effects and Human Health 73

Conclusions on Effects Levels 73

Effects on Populations and Communities from Chronic Exposure to Ionizing Radiation 74

Value of Single-Species Toxicity Tests in the Evaluation of Effects on Populations and Communities 74

Effects of Ionizing Radiation on Populations of Aquatic Organisms 78

Provisional Dose Assessments for Deep-Sea Animals 81

Additional Methodologies for the Study of Radiobiological Effects . . . 84

Monitoring Approaches 85

Recommendations for Future Research on Biological Effects and Biomonitoring Strategies 87

Acknowledgments 89

References and Bibliography 90

Appendix I. List of Major Review Articles 120

Appendix II. Reference Summary Table 121

LIST OF TABLES

1. Induction of mortality in fish from exposure to acute levels of radiation. 13

2. Induction of mortality in invertebrates from exposure to acute levels of radiation. 21

3. Induction of mortality in fish from chronic exposure to radiation. 25

4. Induction of mortality in invertebrates from chronic exposure to radiation. 26

5. Induction of pathophysiological changes in fish from exposure to acute levels of radiation. 27

6. Induction of pathophysiological changes in invertebrates from exposure to acute levels of radiation. 33

7. Induction of pathophysiological changes in fish from chronic exposure to radiation. 34

8. Induction of reproductive changes in fish from exposure to acute levels of radiation. 36

9. Induction of reproductive changes in invertebrates from exposure to acute levels of radiation. 38

10. Induction of reproductive changes in fish from chronic exposure to radiation. 40

11. Induction of reproductive changes in invertebrates from chronic exposure to radiation. 41

12. Induction of developmental changes in fish from exposure to acute levels of radiation. 45

13. Induction of developmental changes in fish from chronic exposure to radiation. 48

14. Induction of developmental changes in invertebrates from exposure to acute levels of radiation. 51

15. Induction of developmental changes in invertebrates from chronic exposure to radiation. 52

16. Induction of chromosomal aberrations in fish from acute and chronic exposure to radiation. 58

17. Induction of mutations in fish from acute and chronic exposure to radiation. 61

18. Induction of chromosomal aberrations in invertebrates from acute and chronic exposure to radiation 64

19. Induction of mutations in invertebrates from acute exposure to radiation. 66

20. Summary of biological effects observed at low dose rates and low total doses in aquatic organisms as compared to selected data for terrestrial organisms. 70

21. Induction of effects on populations of aquatic organisms from chronic exposure to radiation 79

22. Estimates of the radiation dose rates (nSv/h) to benthic deep-sea organisms from natural background and the peak dose rates predicted from dumping low-level radioactive wastes 82

LIST OF FIGURES

1. Stages in the development and modification of radiation injury 5

2. Important endpoints in cytogenetic research and their implications. 56

3. Comparison of yields of dicentrics and centric rings in fish (*A. splendens*, Δ), toad (*B. marinus*, ◇), and man (*H. sapiens* ○) (Woodhead, 1976) 59

4. Generalized effect levels for selected biological endpoints . . . 68

5. The relationship between error of effect-level estimates and stages of investigation (Cairns, 1983). 75

6. Ambient toxicity correlation between *Ceriodaphnia* young per female and ecological survey data (Mount *et al*., 1984) . . . 77

INTRODUCTION

Safe disposal of wastes, whether nuclear or nonnuclear, is the ultimate concern of effective waste management. Wastes presently discharged directly into U.S. marine waters are conservatively estimated to exceed 50 million tons per year. Of this amount, approximately 80% consists of dredged materials, 10% consists of industrial wastes, 9% consists of sewage sludge, and the remaining 1% consists of miscellaneous wastes (Bierman _et al_., 1984). Because the total quantity of these wastes is increasing as the result of increases in population, consideration is being given to alternative disposal strategies and sites.

In the U.S., packaged, low-level, solid radioactive wastes were disposed of off both the Atlantic and Pacific coasts (Joseph _et al_., 1971). Since the onset of the nuclear age, radioactive wastes have been disposed of on land and in the ocean. In addition, the testing of nuclear weapons has contributed measurable quantities of radionuclides to the ocean. Although these practices were discontinued by 1970, little effort was made until recently to determine the subsequent fate and distribution of the radionuclides in these wastes. Information now available indicates that some man-made radionuclides from ocean disposal are present in the bottom sediments, but there is little or no accumulation by organisms in the human food chain (Dyer, 1976; Noshkin _et al_., 1978).

When reviewing requests for permits, U.S. Environmental Protection Agency (EPA) administrators are required to (1) determine that ocean "dumping will not unreasonably degrade or endanger human health, welfare, or amenities, or the marine environment, ecological systems, or economic potentialities" and (2) establish regulations and criteria to implement a permit program (Marine Protection, Research, and Sanctuaries Act, 1972). In 1977, the EPA published final regulations and criteria for ocean dumping. The regulations define the types of environmental information that will be required by EPA to determine criteria for ocean disposal. Evaluation of the potential environmental hazard from a waste-disposal operation involves combining information on the potential biological effects of wastes at given concentrations and knowledge of the fate of the waste material in the receiving water. Fate must be studied to predict the partitioning of a waste material in the environment and to predict the area impacted by disposal operations. Biological effects must be studied to determine the contaminant concentrations constituting threshold-effect levels (level below which lesions are repaired or not realized at the organismal level and no detrimental effects observed).

A problem encountered in defining biological effects of radioactive waste is that traditional bioassays that use mortality as an endpoint are unsuitable for assessing the sublethal effects that may be expected from ocean disposal of low-level radioactive waste. Most mortality tests are limited to observations over a short time period from which long-term assessments of impact are to be inferred. This extrapolation of short-term results to a long-term impact is highly risky because the response elicited from high levels of radiation is different from that to low levels of radiation. Furthermore, many of the species that have been selected for standard testing are inappropriate for evaluating the effects on biota indigenous to the sites proposed for radioactive waste disposal. It is also difficult to determine the uncertainty associated with estimates of effects from exposure to low levels of radiation, because little is known about how specific estimates are modified by changes in experimental parameters such as temperature.

The purpose of this document is to summarize the biological effects of radiation exposure on aquatic organisms and to describe state-of-the-art methods for assessing those effects. Several comprehensive reviews of published data on the effects of radiation on aquatic organisms have been published (Polikarpov, 1966; Templeton et al., 1971; Chipman, 1972; Ophel, 1976; Blaylock and Trabalka, 1978; Egami and Ijiri, 1979; Woodhead, 1984). Full bibliographic information on these review papers is given in Appendix I. Consequently, it is not necessary to give a detailed account of the entire literature. We will synthesize the conclusions on effect levels and generalizations on approaches developed by these authors and provide data relevant to the concern of the Office of Radiation Programs of the EPA, i.e., biological effects that might be expected from the disposal of low-level radioactive wastes into marine environments.

The organization of the report is as follows. First, we discuss the stages in development and modification of radiation injury and explain some differences between the principles of human radiation protection and those for the protection of aquatic life. Then, we discuss effects of acute and chronic exposure to radiation on fishes and aquatic invertebrates. The biological endpoints considered are mortality, pathophysiological changes, reproductive effects, developmental effects, and genetic effects. The endpoints are discussed with respect to their sensitivity and factors that may modify the response. For each endpoint, selected results on terrestrial organisms are provided for comparison of the relative sensitivities of terrestrial and aquatic organisms. Tables provide representative data for each effect category. Critical comments are made, where appropriate, only for the studies demonstrating very low levels of observable effects. Next, population-level studies are analyzed, and the value of single-species toxicity tests in the evaluation of effects on populations and communities is discussed. Then, additional methodologies for the study of radiobiological effects are described, as are monitoring approaches. Finally, recommendations for future research are presented. A table providing the kinds of information included in a group of references reviewed for this report is given in Appendix II. All of these articles are listed in the references, but not all are cited in the text. Hence, the reference list is also a bibliography for which annotations are given in Appendix II.

Because evaluation of the biological effects of radiation exposure is our primary goal, we have considered discussions of the fate of radionuclides in the ocean to be beyond the scope of this work. Hence, discussion of radiation regimes in the ocean is minimal, and representative dose rates are only given for general comparison of effect levels. In a final analysis of effect levels, biological responses to radioactivity from both internal and external emitters and the relative biological effectiveness of the specific radionuclides in the waste in question must be considered. Radiation units used, in citing previously published work, are those of the author(s). In all other instances, the international units, gray (1 Gy = 100 rad) and Sievert (1 Sv = 100 rem), are used. No attempt has been made to formulate a stringent definition of acute versus chronic exposure or high versus low dose rates. Effects of radiation exposure on marine bacteria and plants are not discussed.

STAGES IN THE DEVELOPMENT AND MODIFICATION OF RADIATION INJURY

Before specific effects of radiation exposure in fishes and invertebrates are discussed, we will describe and explain the stages in the development of radiation injury and the nature of radiation syndromes. The types of questions that must be considered in an analysis of radiation effects are:

- What biological molecules or structures are damaged by radiation?
- What types of injury occur, and how are they produced?
- What time interval is required for the effects of radiation to be realized?
- What effects occur at high (potentially lethal) doses versus low (sublethal) doses?
- At what levels of biological organization do different factors that modify the effects of radiation act?

Because these topics are explained in detail in texts on radiation biology such as Arena (1971), only a brief discussion of these topics are given here.

DEVELOPMENT OF RADIATION INJURY

The development of radiation injury occurs at three levels of biological organization: the molecular level, the cellular level, and the organismal level (Fig. 1). When an organism has been exposed to ionizing radiation, energy absorbed in tissues induces atomic changes. These changes may occur through ionization or excitation. Free radicals are produced through ionization, and other types of molecular configurations are produced through excitation. Consequently, molecular effects of ionizing radiation may occur through direct ionization or excitation of an important molecule (e.g., DNA or critical enzymes), or effects may be produced indirectly when the molecule of interest receives energy by transfer from another molecule or is acted on by free radicals (Arena, 1971). When molecular changes are induced in DNA, single- and double-strand breaks result in such effects as mutation, chromosomal breakage, chromosomal rearrangement, and potentially, point mutations (Guerrero et al., 1984). These effects may occur in somatic or germ cells. When they occur in germ cells, effects are sometimes heritable. When molecules other than DNA are damaged, cell death may also occur due to such factors as membrane damage and enzymatic breakdown of cellular structures.

The types of damage realized at the cellular and organismal levels depend on the absorbed dose. For mammals, the causes of death from acute radiation exposure are described below. For massive doses of 100,000 rad (1000 Gy) or more, death occurs during irradiation or immediately thereafter because of inactivation of important biological molecules causing "molecular death." Doses of approximately 10,000 rads (1000 Gy) cause death within a day or two

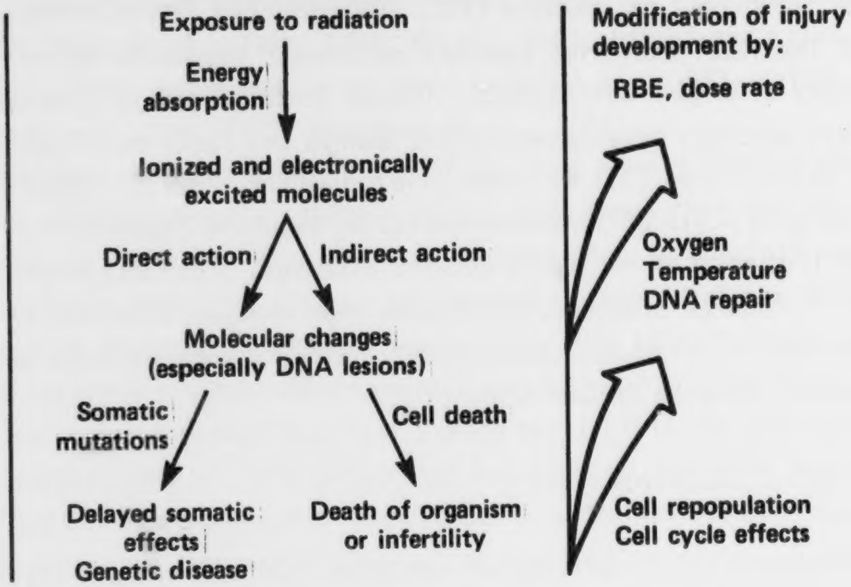

Exposure to radiation
Energy absorption
Ionized and electronically excited molecules
Direct action
Indirect action
Molecular changes (especially DNA lesions)
Somatic mutations
Cell death
Delayed somatic effects
Genetic disease
Death of organism or infertility
Modification of injury development by:
RBE, dose rate
Oxygen
Temperature
DNA repair
Cell repopulation
Cell cycle effects

Figure 1. Stages in the development and modification of radiation injury.

due to central nervous system damage. Between 1000 and 10,000 rads (10 and 100 Gy), death generally occurs in 3 to 5 d or less from damage to cells in the gastrointestinal tract. Loss of these cells and resultant changes in permeability of the gut cause severe imbalance of fluids and electrolytes and bacteremia, which results in death. Doses of 300 to 900 rads (3-9 Gy) cause cell death in blood-forming (hematopoietic) organs. When death occurs at these doses, it is often linked to either secondary infections because of decreased immunity or hemorrhage from loss of platelets (Casarett, 1968).

The major somatic and heritable effects of low-level radiation exposure have been identified for mammals (NRC, 1980; UNSCEAR, 1977). The principal effect of low-level radiation exposure on somatic tissue is cancer induction in a variety of organs and tissues. Oocyte death (possibly from membrane damage) and abnormal development of the embryo and fetus have also been observed below 10 rad (0.1 Gy) (NRC, 1980; UNSCEAR, 1977). Concern over effects on germ cells of low-level radiation exposure centers on induction of increased frequency of heritable genetic diseases. This may occur from mutation, chromosome breakage, chromosome rearrangement, and faulty segregation of chromosomes at metaphase (aneuploidy). Effects such as these may require from hours to years to become evident.

MODIFICATION OF RADIATION EFFECTS

Modification of the effects of radiation can occur at all stages presented in Fig. 1. It is known that the relative biological effectiveness (RBE) of a radiation type has a large effect on the amount of damage produced. Concerning the type of radiation and energy, the most important property affecting RBE is the linear energy transfer (LET). LET is defined as the amount of energy (in keV) dissipated by an ionizing particle per micrometer of path (Arena, 1971). Because of differences in ionization density, the following general hierarchy of biological effectiveness has been established: alpha particles > protons = neutrons > beta particles = gamma rays (ICRP, 1979).

Many responses to radiation exposure are modified by dose rate; a higher total dose is required at low dose rates to produce the same effect observed at high dose rates. This factor has been discussed in detail by Bedford (1982). Because dose rate is an important modifying factor, in this review we will consider chronic, low-level radiation effects separately from those produced as a result of acute exposure.

DNA repair operates at the molecular level and is an important modifier of radiation response. This factor may have particular significance at low dose rates. It is widely debated whether threshold dose rates of radiation exist. However, some repair pathways are known to be error prone and, thus, repair of a lesion does not necessarily imply return to status quo (Schendel, 1981). Furthermore, it is possible that DNA repair enzymes are induced during low-level, chronic exposures (Tuschl et al., 1983). The relative abilities of eukaryotic organisms to repair DNA are unknown. Little is known about DNA repair in aquatic invertebrates (Ejima and Shiroya, 1982), but certain DNA-repair pathways have been examined in fish (Achey et al., 1979; Ishikawa et al., 1978; Mano et al., 1980, 1982; Mitani, 1983; Mitani and Egami, 1982; Mitani et al., 1982; Regan and Cook, 1967; Regan et al., 1983; Walton et al., 1983; Woodhead and Achey, 1979; Woodhead et al., 1978).

Tissue-oxygen concentration is one of the most widely studied modifiers of radiation effects (McNally, 1982). It has been shown that the radioresistance of most systems increases approximately two to fourfold under anoxic conditions. These modifying effects are probably produced at the molecular level. Theories proposed to explain the relationship between oxygen tension and radiosensitivity are that fewer peroxyl and hydroperoxyl radicals are formed at low oxygen tensions and that oxygen may inhibit the DNA repair process (Casarett, 1968). This factor has not been studied with any aquatic organism despite its demonstrated importance in mammalian systems and its possible relevance to deep-ocean disposal of radioactive waste. Oxygen tensions vary greatly between deepwater sites, and this could modify many effects such as gamete death and chromosomal damage.

The effect of temperature on the modification of radiation response occurs at the molecular and cellular levels. Cold temperature is known to slow the development of lethal biochemical lesions and lengthen cell-cycle times, but it may also slow repair processes. The effect of temperature are discussed in more detail in the following section on mortality.

Radiation response is known to depend on the stage in cycling of the cell and on the rate at which cycling is occurring. Cell survival is known to be affected by the stage that is irradiated; S phase, the period in which DNA synthesis occurs, generally is the most radioresistant (McNally, 1982). Types and amounts of chromosomal aberrations induced by radiation also vary with cell-cycle stage at radiation (Wolff, 1968). Moreover, one of the basic tenets of radiation biology is the law of Bergonie and Tribondeau, which states that, in general, the radiosensitivity of cells is directly proportional to their rate of division and inversely proportional to their degree of differentiation. In other words, cell killing occurs selectively in cells that are rapidly dividing and not highly differentiated. This is because one of the prime mechanisms for cell killing is the induction of chromosomal aberrations that cause changes in the genetic complement and in gene expression in the damaged cell. Cell death from aberrations is also due to mechanical interference of the migration of the chromosomes during cell division. Many of these effects will not be realized until the cell divides, and thus cells that are cycling are more sensitive than those that are not. Furthermore, the main reason that relatively less cell killing occurs in highly differentiated cells is that they rarely or never divide. These principles can be applied to radiation effects on aquatic organisms, and they imply that life stage and growth rate of an organism may have a tremendous effect on its radiosensitivity.

When cells are damaged by radiation, they may be replaced by normal cells. This effect is termed cell repopulation, and it is an important modifier of the apparent radiation response at the organismal level. The practical extension of this principle is that when an organism or tissue has the ability to replace damaged cells, effects of radiation at the cellular level will not be realized at the organismal level unless they are in a specialized tissue that cannot be repopulated or unless repopulation occurs slowly. Therefore, the amount of tissue specialization in an organism and/or the ability of tissues to differentiate and dedifferentiate may modify radiation response at the organismal level.

Finally, a distinction on the radiosensitivity of dividing and non-dividing cells must be made. Rapidly dividing cells are most sensitive to acute doses of radiation because of increased probability of cell death. However, slowly dividing cells may be most sensitive to chronic irradiation because they may accumulate chromosomal damage from integration of the dose. Thus, to assess effects in cells exposed to low, chronic doses of radiation for dosimetry or monitoring purposes, it is best to select a non-dividing cell system and then induce the cells to divide so that the damage can be assessed. Effects on DNA in non-dividing cells can also be studied using methods that do not require analysis of metaphase cells, such as quantification of unscheduled DNA synthesis.

PRINCIPLES OF HUMAN RADIATION PROTECTION AND THEIR RELATION TO AQUATIC TOXICOLOGY

Radiation standards to protect humans were developed for different concerns than those to protect aquatic life. The primary concern for human populations was to protect individuals, their progeny, and mankind as a whole against cancer and genetic diseases. The primary concern for aquatic life was to protect ecosystems and their economic potential by maintaining populations of the species indigenous to those ecosystems. Some basic principles of human radiation protection are presented here, and they are discussed as they relate to the approaches used to protect aquatic organisms. The intention of this discussion is to provide a link between the human-radiation-biology community and the aquatic-toxicology community.

In human radiation protection, a distinction is made between stochastic and non-stochastic effects. These are defined as follows (ICRP, 1979):

> "Stochastic" effects are those for which the probability of an effect occurring, rather than its severity, is regarded as a function of dose, without threshold. "Non-stochastic" effects are those for which the severity of the effect varies with the dose, and for which a threshold may therefore occur. At the dose range involved in radiation protection, hereditary effects are regarded as being stochastic. Some somatic effects are stochastic; of these, carcinogenesis is considered to be the chief somatic risk of irradiation at low doses and therefore the main problem in radiation protection.

The goals of human radiation protection are (1) to prevent non-stochastic effects by setting limits at sufficiently low values so that no threshold dose would be reached and (2) to limit the probability of stochastic effects by keeping all justifiable exposures as low as is reasonably achievable; achievement of the latter can be assisted through risk assessment.

Effects on health may be caused by stochastic and non-stochastic effects in exposed individuals and stochastic effects in subsequent generations. The detriment in a population is the expectation of harm based on the probability and severity of the effect. The detriment to health, G, in a group of P persons is given by

$$G = P \sum_i p_i \, g_i$$

where p_i is the probability of suffering the effect i, and g_i is a weighting factor for the severity of the effect (ICRP, 1977).

To predict the probability of suffering a stochastic effect, linear extrapolation is used, based on a cautious assumption of proportionality. For non-stochastic effects, the probability of suffering a given effect is determined by the shape of dose-response curves and threshold doses. The fact that many effects are age and tissue specific is also considered, as are the characteristics of the exposure.

In aquatic toxicology, little is known about frequencies and significance of stochastic effects such as cancer induction and hereditary change. Information to protect aquatic ecosystems is obtained by examining primarily non-stochastic effects. Bioassays to assess short-term changes in mortality rate in different life-history stages or changes in reproductive success are performed. In most cases, the shape of dose-response curves for specific effects have not been well characterized. Consequently, it is not unusual for those in the regulatory community to estimate application factors by such methods as the formulation of acute:chronic ratios (Roop and Hunsaker, 1985). Hence, protection of aquatic life is usually attempted by combining results of standard bioassays with estimates of necessary safety factors. Biomonitoring is sometimes implemented to determine the adequacy of predictions formulated from toxicity testing.

Because the assumption of linearity used in human radiation protection for stochastic effects renders the term "No Observable Effect Level (NOEL)" used in aquatic toxicology unacceptable, and because we know so little about the shapes of dose-response curves for relevant endpoints, we refer to low-effect levels in this report. In a few cases when general aquatic toxicology is discussed, we use the term NOEL as it would be used in that discipline. The pathophysiologic, reproductive, and developmental effects discussed in this report are all considered non-stochastic effects. Only a few studies discussed in the section on genetic effects provide information on stochastic effects.

EFFECTS ON AQUATIC ANIMALS OF ACUTE AND CHRONIC EXPOSURE TO IONIZING RADIATION

MORTALITY

In aquatic toxicology, acute lethal levels of toxicants are usually determined for three purposes. First, they are used to determine relative sensitivities of organisms to lethal effects of a toxic agent. Second, they are used to specify maximum levels of a toxicant permitted for short times in a given environment. Third, they are used to set permissible chronic-exposure levels. The third purpose is addressed in this section. Lethal doses of acute ionizing radiation have been determined for a variety of fishes and aquatic invertebrates. However, information on lethality alone is of limited use in estimating biological effect levels of chronic, low-level, ionizing radiation. Reasons for this conclusion are given in this and subsequent sections.

When lethal levels of a toxicant are established for the purpose of setting maximum chronic exposure levels, safety factors termed "application factors" are used to set a "safe" value below the acute lethality limit. Determination of an appropriate application factor is often subjective. Therefore, it is important to understand the role of factors that modify the level at which mortality will be observed so that an indication of the potential error associated with a predicted safe value can be assessed.

In the section that follows, the criteria for determining a lethal dose are discussed. Next, representative mortality data for various species of aquatic invertebrates and fishes are presented. These are discussed with regard to species specificity and other factors that may modify the levels at which lethal effects are observed.

Criteria for Establishing a Lethal Dose

The LD_{50} is the dose that kills 50% of the organisms tested in a specified amount of time. In acute radiation studies, the dose is given over a short time period of seconds or minutes, and an observation period is specified. In mammalian studies, the standard observation time is 30 or 60 d. Therefore, a lethal dose criterion termed the $LD_{50/30}$ or $LD_{50/60}$ is established.

In poikilotherms (animals, such as fishes and invertebrates, that do not maintain a constant internal body temperature), cell-cycle times are generally much longer and more variable than in mammals. This means that certain types of radiation damage will be realized more slowly in poikilotherms than in mammals. This does not necessarily mean that the cells involved are more radioresistant. Consequently, a 30- to 60-d observation period is generally inadequate. However, there is no standard time period that is suitable for all poikilotherms, and this accounts for the large differences in observation times for the experimental data reported.

Mortality of Fish from Acute Radiation Exposure

Results given in Table 1 demonstrate that the range of lethal levels for adult fish is 375 to 55,000 rad and that 50% mortality of fish embryos has been demonstrated as low as 16 R. Thus, the range of existing data on fish LD_{50} is over 3 orders of magnitude. Modifying factors that have been examined include temperature, salinity, lifestage, RBE, dose rate, and species specificity. However, the role of many modifying factors is still poorly defined, and few studies on marine fish exist. Data provided in Table 1 represent the majority of the information in the literature on fish mortality from acute ionizing radiation.

One of the most important factors affecting LD_{50} values is the experiment duration. It has already been mentioned that 30 d is inadequate for most poikilotherms. However, most of the studies in the literature (Table 1) were terminated at 30 d. Shechmeister *et al*. (1962) demonstrated a dose-dependent decrease in lifespan of the goldfish *Carassius auratus* from 100 to 10,000 rad. These results demonstrate that lethal effects of acute irradiation can be observed at increasingly lower doses, the longer the organism is observed.

In five studies cited in Table 1 (Blaylock and Mitchell, 1969; Egami, 1969a; Etoh and Egami, 1967; Hyodo, 1965a; Lockner *et al*., 1972), the role of temperature in modifying the lethal response to radiation was investigated. These authors concur that low temperature slows the rate of development of lethal lesions and thus postpones death. However, if the observation period is sufficiently long, increased death rates from low doses may still be detected. A finding of particular interest is that low temperature may negate dose-rate effects; this implies that accumulation or integration of effects at

Table 1. Induction of mortality in fish from exposure to acute levels of radiation.

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
16 R (50%, 150 d)	*Oncorhynchus kisutch*[c] (silver salmon)/1-cell stage	x ray 12-2400 R	--	Bonham and Welander, 1963
58 R (50%, 55 d)	*Salmo gairdnerii*[c] (rainbow trout)/1-cell stage embryos	x ray 25-2570 R	One-cell or blastodisc stage most sensitive	Welander, 1954
90 rad (50%, blastula to metamorphosis)	*Pleuronectes platessa*[d] (plaice)/embryos	x ray 30-150 rad	LD_{50} at metamorphosis for plaice larvae irradiated at blastula stage	Ward *et al.*, 1971
300-500 R (50%, 55 d)	*Salmo gairdnerii*[c] (rainbow trout)/32-cell to late eye embryos	x ray 25-2570 R	Irregular relationship of sensitivity and increased dose	Welander, 1954
375 rad-neutron (50%, 30 d)	*Carassius auratus*[c] (goldfish)/adults	Fast neutrons, 2 MeV 40 rad/min 250-3000 rad	Relative biological effectiveness (RBE) discussed	Etoh *et al.*, 1974
1283 rad-x ray (50%, 30 d)		x ray 475-1663 rad		
1000 rad (50%, 60 d) 2500 rad (50%, 30 d)	*Carassius auratus*[c] (goldfish)/adults	x ray 100-10,000 rad	Dose dependent decrease in lifespan observed from 100-10,000 rad	Shechmeister *et al.*, 1962
1020 rad (25 ppt)[e] 1540 rad (15 ppt) 2050 rad (5 ppt) (50%, 30 d)	*Fundulus heteroclitus*[d] (mummichog)/adults	^{60}Co 365 rad/min 500-2000 rad	Increased sensitivity with increased salinity and temperature	Angelovic *et al.*, 1969

Table 1. (Continued.)

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
1050 R (50%, 30 d)	*Micropogon undulatus*-- (--)/juveniles or postlarvae	^{60}Co -- --	--	White and Angelovic, 1966 (in Chipman, 1972)
1120 R (50%, 30 d)	*Fundulus heteroclitus*[d] (mummichog)/juveniles or postlarvae	^{60}Co -- --	--	White and Angelovic, 1966 (in Chipman, 1972)
1200 rad (50%, 30 d, 18°C) 55,000 rad (50%, 30 d, 10°C)	*Tinca vulgaris*[c] (tench)/adults	^{60}Co 2 rad/s (18°C) and 250 rad/s (10°C) 0-60,000 rad	Note different dose rates and short time period (30 d); temperature effects observed	Lockner *et al.*, 1972
1450 R (50%, 30 d)	*Mugil cephalus*[c] (mullet)/juveniles or postlarvae	^{60}Co -- --	--	White and Angelovic, 1966 (in Chipman, 1972)
2350 R (50%, 30 d)	*Poecilia reticulata*[c] (guppy)/1-week old	x ray --	--	Erickson, 1973
2500 R (50%, 60 d)	*Oncorhynchus tschawytscha*[c] (chinook salmon)/embryos	x ray 250-10,000 R	No significant mortality at 30 d; however, 50% mortality was obtained in 40 d in all doses $\geq$ 2500 R; significant increases in mortality occurred at 1000 R	Welander *et al.*, 1948

Table 1. (Continued.)

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
3000 R (50%, 30 d)	*Lagodon rhomboides*[c] (pinfish)/juveniles or postlarvae	^{60}Co -- --	--	White and Angelovic, 1966 (in Chipman, 1972)
3500 R (50%, 30 d)	*Eucinostomus sp.*[d] (--)/juveniles or postlarvae	^{60}Co -- --	$LD_{50/50}$ was 2000 R	White and Angelovic, 1966 (in Templeton *et al.*, 1971)
3699 rad (50%, 30 d)	*Gambusia affinis affinis*[c] (mosquitofish)/adults	^{60}Co 8.2 rad/sec 1500-6000 rad	Tests at 20, 25, and 30°C showed that sensitivity increased with increased temperature; no evidence of increased radioresistance of population exposed chronically for 10 generations to $\leq$10 rad/d	Blaylock and Mitchell, 1969
5000 R (100%, 7 d)	*Lagodon rhomboides*[c] (pinfish)/adults	Gamma rays 385 R/h 5000 R	Changes in cellular components of blood described	Engel *et al.*, 1967
5550 R (50%, 30 d)	*Paralichthys lethostigma*[d] (plaice)/juveniles or postlarvae	^{60}Co -- --	$LD_{50/50}$ was 2500 R	White and Angelovic, 1966 (in Templeton *et al.*, 1971)
8000 rad (100%, 30 d, 22°C) 8000 rad 83%, 30 d, 4°C)	*Carassius auratus*[c] (goldfish)/adults	x ray 0, 8000 rad	Low temperature inhibits the development of intestinal damage, which would lead to death	Hyodo, 1965a

Table 1. (Continued.)

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
10,000 R (41.7%, 30 d)	*Carassius auratus*[c] (goldfish)/adults	x ray 1000-16,000 R	Mortality rate following partial-body irradiation depended on exposure and damage of the directly irradiated part, as well as absorbed dose in the tissue as a whole	Etoh *et al.*, 1968
--	*Pimephales promelas*[c] (fathead minnow)/ adults	^{60}Co 200-3000 rad at 2.62 rad/min 600-6000 rad at 37.8 rad/min 2300-24,000 rad at 1730 rad/min	Time to 50% mortality: 3000 rad (2.62 rad/min) - 760 d (37.8 rad/min) - 28 d (1730 rad/min) - 17 d	Chee *et al.*, 1979
--	*Oryzias latipes*[c] (medaka)/adults	x ray Single dose 100,000 R or 2 X 50,000 R. Single dose 4000 R or 2 X 2000 R and 2000-4000 R fractionated	If interval between fractionations is 3 d or longer, mortality from radiation was reduced	Egami and Etoh, 1966
--	*Oryzias latipes*[c] (medaka)/adults	x ray 1000-31,000 R (includes numerous fractionations)	Longer intervals between fractionations diminished the effect of irradiation; decreased effects due to fractionation did not occur at low temperatures	Etoh and Egami, 1967
--	*Oryzias latipes*[c] (medaka)/adults	^{60}Co 14 R/min--1000-4000 R 0.6, 0.8, 5.4, 10.5, and 105 R/min--3600-4800 R	Dose-rate effect observed but negated at low temperature (6°C) (0% survival at 3000-6000 R over 4 d at 6°C vs 80% at 25°C)	Etoh and Egami, 1967

Table 1. (Continued.)

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
--	*Oryzias latipes*[c] (medaka)/adults	x ray Many split doses	When 3000-R dose was split in half with 3 d between doses, 10% (24°C) and 40% (15°C) mortality were observed at 30 d, vs 80% (24°C and 15°C) mortality when the dose was delivered continuously	Egami, 1969a

---

[a] Lethal dose is expressed as the dose that caused a specified percent mortality in a specified number of days. Units are those used by the authors, R is the abbreviation for roentgens.

[b] Radiation regime is presented as source, dose rate, and total dose; a dashed line indicates information was not available. For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[c] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[d] Organism is exclusively marine.

[e] ppt is the abbreviation for parts per thousand.

low dose rates may occur (Etoh and Egami, 1967). Possible negation of dose-rate effects may introduce large uncertainties in predictions of biological effect levels in organisms in the deep ocean from data available on coastal organisms. Accumulation of chromosomal damage from chronic exposure could result in unusually high effects at low dose rates, but slowing of lesion development and cell cycling due to cold temperature could impart tremendous radioresistance. It is not known what the relative importance of these factors would be.

Effect of low temperature on radiation injury has been examined in animals other than fishes and aquatic invertebrates because biomedical researchers have been interested in the role of metabolic rate on the development of radiation-induced, molecular lesions. Some early findings were summarized in Bacq and Alexander (1961), as follows:

> If frogs irradiated with 3000 to 6000 r at 23°C are cooled to 5°C, 80 to 90 percent of the animals survive for more than 3 to 4 months after irradiation, whereas controls kept at 23°C die in 3 to 6 weeks. The lesion in the cooled animals is latent and if they are warmed after 60 to 130 d they die. No lesion is found in the ovarian ova of amphibians 12 d after total irradiation with 3000 to 5000 r if the animals are kept at 5°C, but if they are kept at 22°C after exposure to 3000 r, all the ova are affected. If the chilled and irradiated animals are warmed after 12 d the lesions appear very quickly.
>
> Lamarque and Gros irradiated the eggs of the silk worm (Bombyx mori) and kept them in the cold; when they were warmed six months later, it was found that very few of the radiation lesions had been repaired.
>
> Similarly, hibernating mammals such as squirrels or marmots are much less radiosensitive when irradiated and maintained in hibernation. On warming 2 to 4 weeks after irradiation the animals show the radiation response of animals exposed to x rays in the non-hibernating state and die in about 20 d following a lethal dose.

A logical extension of the observations of Bacq and Alexander (1961) is that organisms that receive a radiation dose in cold deep water and then migrate to warm surface waters could be at increased risk. More recently, cell kinetics and cell survival at lowered temperatures have been examined in detail, and further evidence for increased radiosensitivity was obtained (e.g., Van Rijn et al., 1985; and Szechter and Schwarz, 1977).

Salinity may also modify the level at which lethal effects of radiation are observed. Angelovic et al. (1969) found an $LD_{50/30}$ for the mummichog Fundulus heteroclitus of 2050 rad at a salinity of 5 ppt and 1020 rad at 25 ppt.

It has been widely established (Ophel, 1976; Donaldson and Foster, 1957) that radiosensitivity varies with age and that early lifestages are more sensitive than adults. LD_{50} values below 100 R have been reported for a variety of fish species. They include 16-R $LD_{50/30}$ for single-cell embryos of the silver salmon Oncorhynchus kisutch (Bonham and Welander, 1963), 90-rad LD_{50} for survival to metamorphosis in larvae of the plaice Pleuronectes platessa (Ward et al., 1971), and 58-R $LD_{50/55}$ for the rainbow trout Salmo gairdnerii irradiated as single-cell embryos (Welander, 1954). These values are low relative to the LD_{50} values of above 1000 rad obtained for adult fish in most of the studies reported in Table 1. Thus, the lethal effects of radiation in fishes are highly dependent on lifestage at irradiation, and differences in LD_{50} of more than two orders of magnitude may be expected for different stages of the same species.

The relative biological effectiveness (RBE) of the radiation and dose rate delivered by a radiation source may also alter the response. A high RBE for neutrons in mammals has been demonstrated (Casarett, 1968). Similarly, Etoh et al. (1974) found that neutrons were four times more effective than x rays in the induction of mortality in goldfish.

For a given total dose, the effects of decreasing the dose rate and fractionating the dose have been studied in fish (Chee et al., 1979; Egami, 1969a; Egami and Etoh, 1966; Etoh and Egami, 1967). Classic results, similar to those in mammals, have been obtained, i.e., as dose rate is decreased and as intervals between fractionated doses are increased, a greater total dose is required to produce the same biological effects observed at high dose rates with no fractionation.

Species-specific differences in radiation-induced lethality are difficult to evaluate from the data available (Table 1) because of the differences among studies in exposure conditions, lifestages, and observation periods. However, the research of White and Angelovic (1966), reported in Chipman (1972) and Templeton et al. (1971), shows that for six species of marine fish irradiated with ^{60}Co as juveniles or postlarvae, the $LD_{50/30}$ ranged from 1050 R to 5550 R. Juveniles and postlarvae are actively growing organisms, but because not all may be growing at the same rate, differences in sensitivity may be attributable to differences in growth rate as well as to differences in species.

Mortality of Invertebrates from Acute Radiation Exposure

The comparative radiosensitivity of living organisms has been discussed in numerous reviews (e.g., Whicker and Schultz, 1982). Although many factors determined the radiosensitivity of an organism, it has been emphasized that, among other factors, organismal radiosensitivity increases with increasing complexity of the organism. Bacq and Alexander (1961) report $LD_{50/30}$ values for some unicellular protists such as *Paramecium sp.* to be in excess of 100,000 R. $LD_{50/30}$ values for many higher invertebrates (Table 2) range in the thousands and ten thousands of rads (tens and hundreds of Gy). However, values as low as 210 rad (Engel *et al.*, 1974) have been reported. While the role of some modifying factors such as salinity and species specificity have been examined (Engel *et al.*, 1974 and Table 2), the lower limit of radiation inducing mortality in invertebrates is probably still undefined. In particular, very little work has been done to determine the radiosensitivity of gametes and early lifestages of invertebrate species. Other areas of weakness are the short observation times used in many of the studies and the absence of data on temperature effects.

Some work that has been completed on mortality of early lifestages of invertebrates indicates greater radioresistance of embryos of invertebrates than of fish. Ravera (1968), reported in Blaylock and Trabalka (1978), showed LD_{50} values for the four-cell, trochophore, veliger, and hippostage embryos of the gastropod *Physa acuta* to be 1075, 2350, 4900, and 10,750 R, respectively. In contrast, Bacq and Alexander (1961) cite work on *Drosophila* that indicates an LD_{50} for 3-h *Drosophila* embryos of 200 R. It must be re-emphasized at this point that the results of such studies depend not only on the dose received but also on many other inherent experimental parameters such as observation time, mitotic delay at high doses, differences in cell-cycle time, and temperature. Furthermore, similar radiosensitivities of related species cannot be assumed, and rules on increasing radiosensitivity with phylogenetic position are not absolute. Hoppenheit (1969), in Chipman (1972), has shown that the $LD_{50/30}$ of the amphipod *Gammarus duebeni* (3900 R) is twice that of *Gammarus zaddachi* (1700 R). In addition, the $LD_{50/30}$ of the more phylogenetically advanced crustacean *Callinectes sapidus* (blue crabs) is 56,600 R (Engel, 1967, in Chipman, 1972). Differences in radiosensitivity between sexes have also been observed. In the mollusc *Urosalpinx cinerea*, males ($LD_{50/70}$ of 15,000 R) are twice as radiosensitive as females ($LD_{50/70}$ of 30,000 R).

Table 2. Induction of mortality in invertebrates from exposure to acute levels of radiation.

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
210 R (30 ppt)[c] 405 R (25 ppt) 450 R (20 ppt) 600 R (15 ppt) (50%, 40 d)	Palaemonetes pugio[d] (grass shrimp)/adults	-- -- 200-4800 R	Salinity effects observed	Engel et al., 1974
1075 R (50%, --)	Physa acuta[d] (freshwater snail)/ four-cell embryos	--	--	Ravera, 1968 (in Blaylock and Trabalka, 1978)
1700 R (50%, 30 d)	Gammarus salinus[e] (amphipod)/ --	--	--	Hoppenheit, 1969 (in Chipman, 1972)
1700 R (50%, 30 d)	Gammarus zaddachi[e] (amphipod)/ --	--	--	Hoppenheit, 1969 (in Chipman, 1972)
2000-9000 R (100%, 35-75 d)	Hydra fusca[--] (coelenterate)/ --	--	--	Nikitin, 1938 (in Polikarpov, 1966)
3500 R (50%, 30 d)	Gammarus duebeni[e] (amphipod)/females	--	--	Hoppenheit, 1969 (in Chipman, 1972)
3900 R (50%, 30 d)	Gammarus duebeni[e] (amphipod)/males	--	--	Hoppenheit, 1969 (in Chipman, 1972)
5000 R (50%, 80 d)	Crassostrea virginica[e] (oyster)/ --	^{60}Co -- --	--	White and Angelovic, 1966 (in Templeton et al., 1971)

Table 2. (Continued.)

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
5500 R (100%, 75 d)	Planaria polychroa[--] (planarian)/ --	--	--	Nikitin, 1938 (in Polikarpov, 1966)
8000 R (50%, 50 d)	Uca pugnax[e] (fiddler crab)/females	^{60}Co -- --	--	White and Angelovic, 1966 (in Templeton et al., 1971)
10,000 R (100%, 60 d)	Dendrocoelum lacteum[--] (annelid)/ --	--	--	Schmidt, 1946 (in Polikarpov, 1966)
10,000 R (50%, 35 d)	Calliopius laeviusculus[e] (amphipod)/ --	--	--	Bonham and Palumbo, 1951 (in Chipman, 1972)
10,000 R (50%, 35 d)	Allorchestis angustus[e] (amphipod)/ --	--	--	Bonham and Palumbo, 1951 (in Chipman, 1972)
10,000 R (50%, 50 d)	Arbacia punctulata[e] (sea urchin)/ --	--	--	White and Angelovic, 1966 (in Templeton et al., 1971)
12,000 R (100%, 63 d)	Pelmatohydra oligactis[--] (coelenterate)/ --	--	--	Polikarpov, 1957a,b (in Polikarpov, 1966)
20,000 R (50%, 50 d)	Nassarius obsoletus[e] (mollusc)/males	^{60}Co -- --	--	White and Angelovic, 1966 (in Templeton et al., 1971)
28,000 R (50%, 24 d)	Physa acuta[d] (freshwater snail)/adults	x ray 2000-220,000 R	Behavioral changes before death	Ravera, 1967

Table 2. (Continued.)

Lethal dose[a] (% mortality, days)	Organism/lifestage	Radiation regime[b]	Comments	Reference
30,000 R (50%, 50 d)	Urosalpinx cinerea[e] (mollusc)/males	^{60}Co -- --	--	White and Angelovic, 1966 (in Templeton et al., 1971)
40,000 R (50%, 50 d)	Urosalpinx cinerea[e] (mollusc)/ females	^{60}Co -- --	--	White and Angelovic, 1966 (in Templeton et al., 1971)
50,000 rad (50%, 73 d;adults) (50%, 45 d;juveniles)	Neanthes arenaceodentata[e] (polychaete)/adults and juveniles	^{137}Cs 500 rad/min 100-100,000 rad	Effects of cell reproduction were considered	Anderson et al., in prep.
50,000 R (50%, 80 d)	Mercenaria mercenaria[e] (clam)/ --	^{60}Co	--	White and Angelovic, 1966 (in Templeton et al., 1971)
56,600 R (50%, 30 d)	Callinectes sapidus[e] (blue crab)/adults	--	--	Engel, 1967 (in Chipman, 1972)
--	Diaptomus clavipes[d] (calanoid copepod)/ adults and embryos	^{60}Co 7000 rad/min 1000-100,000 rad	Mean survival time was significantly decreased at 1000 rad; lowest dose $LD_{50/30}$ cannot be determined due to short lifespan; control death occurred prior to 30 d	Gehrs et al., 1975

---

[a] Lethal dose is expressed as the dose that caused a specified percent mortality in a specified number of days. Units are those used by the authors; R is the abbreviation for roentgen.

[b] Radiation regime is presented as source, dose rate, and total dose; a dashed line indicates information was not available. For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[c] ppt is the abbreviation for parts per thousand.

[d] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[e] Organism is exclusively marine.

Mortality of Fish and Invertebrates from Chronic Radiation Exposure

In a few studies, mortality in chronically irradiated fish and invertebrates (see Tables 3 and 4) is examined. Donaldson and Bonham (1964) reported no significant differences in mortality between the salmon *Oncorhynchus tschawytscha* embryos irradiated with 0.5 R/d for approximately 20 d and the control salmon embryos; observations were conducted up to the time of release of the smolts. Erickson (1973) reported no increase in mortality of the guppy *Poecilia reticulata* exposed to 0.05 to 1 mCi/mL of tritium (total doses of 340-4700 rad). Adults of the blue crab *Callinectes sapidus* subjected to chronic gamma irradiation required dose rates greater than 29 rad/h for 70 d to cause death (Engel, 1967), and juveniles of the clam *Mercenaria mercenaria* exposed to 0.006-37.0 rad/h for 14 months exhibited only decreases in reproduction and growth at the highest dose rate, 16-37 rad/h (Baptist *et al*., 1976).

Conclusions on Mortality Data

Although it is clear that many aspects of mortality due to radiation injury in aquatic species remain obscure, it is not likely that observable numbers of aquatic organisms will die from the radiation levels that are present at current ocean dumpsites. The highest dose rate observed from the release of low-level, radioactive waste in the ocean is 0.240 rad/d, which occurred at the end of the pipeline at the Windscale reprocessing plant in the U.K. (Woodhead, 1984). Such doses are not expected over large areas at ocean dumpsites (NEA, 1980); however, we use this figure as the documented maximum level for our discussion. At 0.240 rad/d, mortality has not been observed in any aquatic organism (Tables 1-4). However, very few studies at chronic dose rates have been conducted. It is likely that reproductive and genetic effects will have more relevance at these levels. Nevertheless, it is not totally inconceivable that chronic irradiation of sensitive lifestages such as brooded embryos, which may be very slow in developing at cold temperatures, could accumulate sufficient damage to cause death. How, or if, the biochemical lesions would be realized at such low temperatures is not known. In conclusion, the lowest observed lethal effect level is a 16-R $LD_{50/150}$ for single-cell silver salmon embryos (Bonham and Welander, 1963). Methods and

Table 3. Induction of mortality in fish from chronic exposure to radiation.

Organism/lifestage	Radiation regime[a]	Comments	Reference
Oncorhynchus tschawytscha[b] (chinook salmon)/embryos	^{60}Co 0.5 R/d 33-40 R	No significant differences in mortality	Donaldson and Bonham, 1964
Poecilia reticulata[b] (guppy)/young fish	Tritium 0.05-1 mCi/mL 340-4700 rad	No significant increase in mortality	Erickson, 1973

[a] Radiation regime is presented as source, dose rate, and total dose. Units are those used by the author, R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

analyses used in this study are suitable. However, it should be noted that the $LD_{50/150}$ of salmon embryos at slightly different incubation times (or developmental stages) ranged from 16 R to 309 R.

PATHOPHYSIOLOGY

In this section on pathophysiological effects, we describe research conducted on several types of somatic tissue. Research on germ tissue or developing embryos is presented in the sections on reproduction and development.

Pathophysiological Effects in Fish from Acute Radiation Exposure

Research has defined a variety of pathophysiological lesions in fish from acute radiation exposure. In most such studies, endpoints that characterize radiation syndromes in mammals have been examined. These include effects on hemopoietic tissue (Welander *et al.*, 1948; Etoh *et al.*, 1968; Cosgrove *et al.*,

Table 4. Induction of mortality in invertebrates from chronic exposure to radiation.

Organism/lifestage	Radiation regime[a]	Comments	Reference
Callinectes sapidus[b] (blue crab)/adults	^{60}Co 3.2-29.0 rad/h 70 d	Decreased survival only at highest dose rate, 29.0 rad/h	Engel, 1967
Mercenaria mercenaria[b] (clam)/juvenile	^{60}Co 0.006-37.0 rad/h 14 mo	Deleterious effects on growth and survival only at highest dose rate of 16-37 rad/h with only 10% survival to 14 mo	Baptist et al., 1976
Argopecten irradiens[b] (scallop)/juvenile	^{60}Co 0.006-37.0 rad/h 3 mo	No deleterious effects on growth and survival	Baptist et al., 1976

---

[a] Radiation regime is presented as source, dose rate, and total dose. Units are those used by the author.

[b] Organism is exclusively marine.

1975; Lockner et al., 1972), effects on immunity (Preston, 1959; Shechmeister et al., 1962), or damage to the intestinal epithelium (Hyodo, 1965a; Etoh et al., 1968; Hyodo-Taguchi and Egami, 1969; Johnson et al., 1970). However, with the exception of studies relating to immunity, most of these effects have been studied at lethal levels, and they mainly serve to characterize lethal radiation syndromes in fish (Table 5). The few studies that document effects below 1000 rad are discussed below.

Hemopoietic cells of fish are affected by sublethal doses of radiation. Welander et al. (1948) observed a significant decrease in the number of hemopoietic cells in embryos of the chinook salmon Oncorhynchus tschawytscha

Table 5. Induction of pathophysiological changes in fish from exposure to acute levels of radiation.

Endpoint	Organism/lifestage	Radiation regime^a	Comments	Reference
Susceptibility to infection	*Carassius auratus*^{b} (goldfish)/adults	x ray 1000, 2000 rad for *Aeromonas* study 100, 1000 rad for *Gyrodactylus* study	100% mortality was observed at 1000 rad when infected with *Aeromonas salmonicida* compared to 0% in uninfected irradiated (1000 rad) fish and infected unirradiated fish 78% mortality was observed at 100 rad in fish infected with *Gyrodactylus sp*., whereas unirradiated infected fish exhibited approximately 15% mortality (day 20)	Shechmeister *et al*., 1962
Immune response	Plaice^c/--	x ray 8000 R	Irradiated fish could still develop leucocytosis in response to bacteremia; this was demonstrated in two fish that died at 45 and 51 d after exposure	Preston, 1959
Hemopoiesis	*Tinca vulgaris*^{b} (tench)/adults	^{60}Co 2 rad/sec and 250 rad/sec 500-60,000 rad	At 18°C, 100 rad caused 50% decrease in lymphocyte count; at 10°C, 16,000 rad are required for the same effect	Lockner *et al*., 1972
Hemopoiesis	*Ictalurus punctatus*^{b} (channel catfish)/young	^{60}Co 305 rad/min 500-2500 rad	Severe atrophy of hemopoietic tissue in kidney, spleen, and thymus in all fish at 500-2500 rad; recovery of hemopoietic system seen after 29 d	Cosgrove *et al*., 1975
Hemopoiesis	*Gambusia affinis*^{b} (mosquitofish)/adults	^{60}Co 360 rad/min 750-6000 rad	750 rad caused only slight depression of hemopoiesis (endpoint not specified)	Cosgrove *et al*., 1975
Hemopoiesis	*Lepomis macrochirus*^{b} (bluegill)/adults	^{60}Co 160 rad/min, 1000, 3000 rad	1000 rad caused atrophy in the hemopoietic system of kidney within 3 d	Cosgrove *et al*., 1975

Table 5. (Continued.)

Endpoint	Organism/lifestage	Radiation regime[a]	Comments	Reference
Hemopoiesis	*Lagodon rhomboides*[c] (pinfish)/adults	Gamma ray -- 2000 and 5000 R	Number of thrombocytes in irradiated fish decreased linearly with time; rate of decrease was greater in fish irradiated at higher dose; number of leucocytes decreased also	Engel *et al.*, 1967
Hemopoiesis and glomerular development	*Oncorhynchus tschawytscha*[b] (chinook salmon)/ embryos	x ray 250-10,000 R	Number of hemopoietic cells significantly decreased at 500 R while at the same exposure, development of glomeruli was only slightly inhibited	Welander *et al.*, 1948
Concentration of serum-protein fractions	*Lepomis macrochirus*[b] (bluegill)/yearlings	^{60}Co 167 R/min, 1000-3000 R	An initial 50% decrease in beta globulins, alpha globulins, and albumins occurred within 2-24 h after exposure; hemoconcentration occurred after initial decrease	Ulrikson, 1973
Blood coagulation	*Lagodon rhomboides*[c] (pinfish)/adults	x ray 100-1600 R	Increase in the coagulation time with exposures above 100 R	Engel *et al.*, 1965
Blood coagulation	*Fundulus sp.*[c] (mummichog)/adults	x ray 100 R	Increase in coagulation time greatest at 24 h and approached that of controls at 96 h	Engel *et al.*, 1965
Damage to intestinal epithelium	*Oncorhynchus kisutch*[b] (coho salmon)/juveniles	x ray 140-8000 R	At 500 R, minor and temporary effects were observed; doses of 1000 R and above caused considerable cell damage with only partial recovery	Johnson *et al.*, 1970
Damage to intestinal epithelium	*Carassius auratus*[b] (goldfish)/adults	x ray 0, 8000 rad	Fish irradiated at 8000 rad and held at 22°C developed intestinal damage, which lead to death, but those held at 4°C did not develop intestinal damage unless they were later transferred to the warmer water	Hyodo, 1965a

Table 5. (Continued.)

Endpoint	Organism/lifestage	Radiation regime[a]	Comments	Reference
Damage to intestinal epithelium	*Carassius auratus*[b] (goldfish)/adults	x ray 0, 8000 R	Ratio of mitotic figures (^{3}H-thymidine labeled:unlabeled) was lower in irradiated fish intestine when compared to controls	Hyodo, 1965b
Damage to intestinal epithelium	*Oryzias latipes*[b] (medaka)/adults *Carassius auratus*[b] (goldfish)/adults	x ray 50-128,000 R x ray 4000-32,000 R	Pathological changes in intestinal epithelium responsible for acute radiation death at the "dose-independent range" (3000-30,000 R). Mitotic activity of epithelial cells of intestine completely inhibited; response temperature dependent; histological changes in intestine correlated with radiation death	Hyodo-Taguchi and Egami, 1969
Thyroid damage	Various coral reef fishes[c]	Nuclear bomb test at Enewetak Atoll (^{131}I)	Thyroid damage in coral reef fishes from bomb testing at Enewetak Atoll was demonstrated histopathologically	Gorbman, 1963
Thyroid follicle enlargement	*Salmo salar*[b] (Atlantic salmon)/ larvae	x ray 0, 350, 1000 R	Increased size of thyroid gland follicles at 1000 R; a very slight increase was also observed at 350 R	Oganesyan, 1973
Thymus volume	*Oryzias latipes*[b] (medaka)/adults and embryos	Adults: γ ray 50 R/min 200 and 500 R Embryos: γ ray -- 1000-4000 R	Decreased thymus volume in adults showed recovery; decreased thymus volume in embryos showed no recovery at 4000 R	Ghoneum and Egami, 1980

Table 5. (Continued.)

Endpoint	Organism/lifestage	Radiation regime[a]	Comments	Reference
Mitotic delay	Carassius auratus[b] (goldfish)/adults	x ray 1000, 2000, and 8000 R	At 3-6 h after exposure, the number of thymidine-labeled intestinal epithelial cells in the irradiated groups did not differ from controls; after 24-48 h, the number of labeled cells in the 8000-R group approached 0; in the 1000- and 2000-R groups an initial depression occurred with recovery between 48 and 96 h	Hyodo-Taguchi, 1970
Inhibition of DNA synthesis	Oncorhynchus kisutch[b] (coho salmon)/juveniles	x ray 1000, 2000, and 4000 R	At all doses, DNA synthesis was depressed from 10-160 min; no longer time points or lower doses were used	Johnson et al., 1970
Formation of taste bud cells	Oryzias latipes[b] (medaka)/embryos and adults	^{137}Cs 360 R/min (embryos) 250 R/min (adults) 1000 R	Partial inhibition of taste-bud-cell formation in embryos; number of taste-bud cells in adults declined but repopulation appeared to minimize effects	Gnoneum et al., 1983
Damage to skin, intestine, gill, and hemopoietic tissue	Carassius auratus[b] (goldfish)/adults	x ray 1000-16,000 R	Histological damage to intestinal and hemopoietic tissue was demonstrated after 10 and 20 d at 1000 R; irradiation of the tail region showed skin damage to be an important cause of death; irradiation of the head region caused gill damage, which caused sodium loss through damaged tissue	Etoh et al., 1968
Interrenal cell hypertrophy	Carassius auratus[b] (goldfish)/adults	x ray 250-128,000 R	Hypertrophy of interrenal cells was produced in fish at 1000-16,000 R; this effect was not seen in hypophysectomized fish, and it was suggested that ACTH secretion was stimulated by irradiation	Aoki et al., 1966

Table 5. (Continued.)

Endpoint	Organism/lifestage	Radiation regime[a]	Comments	Reference
Osmoregulation	*Oncorhynchus kisutch*[b] (coho salmon)/juveniles	x ray 1000-1200 rad	Osmoregulatory ability decreased at lethal levels: (a) decreased survival of fish at doses greater than 1000 rad when transferred from hypoosmotic to hyperosmotic conditions (when compared to those maintained under hypoosmotic conditions); (b) increased plasma sodium, chloride, and osmotic concentrations in surviving irradiated fish maintained in salt water	Conte, 1965
Osmoregulation	*Anguilla anguilla*[b] (eel)/adults	^{60}Co -- 1000 R	Effect of increased salinity and irradiation caused additive increase in lipid biosynthesis in eel gills (assumes changes in membrane permeability accompanied by changes in the composition of the lipid moiety)	Hansen, 1975
Osmoregulation	*Anguilla anguilla*[b] (eel)/adults	x ray 1000 R	Radiation and salinity effects are decreased *in vitro*, implying neurohormonal regulation of these effects	Hansen, 1980
External morphology	*Eucinostomus sp.*[c] (mojarra)/post-larvae	x ray 400-3200 R	Apparent decreased growth rate with increased exposure; changes in body proportions, size of spines, and pigmentation.	Engel *et al.*, 1965

[a] Radiation regime is presented as source, dose rate, and total dose; a dashed line indicates information was not available. For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Organism is exclusively marine.

after an x-ray exposure of 500 R. Cosgrove et al. (1975) noted severe hemopoietic atrophy in kidney, spleen, and thymus of the channel catfish Ictalurus punctatus after a 500-rad dose. Lockner et al. (1972) observed a 50% decrease in lymphocyte count when the tench Tinca vulgaris received 100 rad at 18°C, whereas at 10°C, 16,000 rad were required to produce the same effect in the specified observation period.

Immunity of irradiated goldfish Carassius auratus was examined by Shechmeister et al. (1962). It was found that fish infected with Gyrodactylus sp. and receiving up to 100 rad from an x-ray source exhibited 78% mortality. In contrast, unirradiated infected fish exhibited only 15% mortality.

Damage to intestinal epithelium is a characteristic acute lethal radiation syndrome in mammals, and, in fishes, it has also been studied mostly at lethal levels. However, minor and temporary damage to the intestinal epithelium of coho salmon Oncorhynchus kisutch juveniles exposed to 500 R from a x-ray source has been documented (Johnson et al., 1970). Exposures of 1000 R and above caused considerable cell damage with only partial recovery.

Pathophysiological Effects in Invertebrates from Acute Radiation Exposure

Little or no work has been done to characterize the pathophysiological effects of acute ionizing radiation on aquatic invertebrates (Table 6). However, Engel et al. (1974) found that irradiation (200 to 4800 rad) and increased salinity acted additively in affecting amino acid pools of the estuarine shrimp Palaemonetes pugio. These results may indicate that irradiation causes membrane damage that inhibits regulation of intracellular amino-acid pools used in osmoregulation.

Pathophysiological Effects in Fish and Invertebrates from Chronic Radiation Exposure

To our knowledge, no research has been conducted that characterizes pathophysiological effects from chronic radiation exposure in aquatic invertebrates. This discussion will, therefore, be restricted to results of research conducted on fish (Table 7).

Cosgrove et al. (1975) examined the mosquitofish Gambusia affinis for hemopoietic damage after exposure to ^{60}Co at 0.5 to 5.43 rad/h for up to 128 d. They found no demonstrable damage to hemopoietic organs after 37 d at

Table 6. Induction of pathophysiological changes in invertebrates from exposure to acute levels of radiation.

Endpoint	Organism/lifestage	Radiation regime[a]	Comments	Reference
Osmoregulation	Palaemonetes pugio[b] (grass shrimp)/ adults	-- -- 200-4800 rad	Radiation and increased salinity act additively, affecting amino acid pools	Engel *et al.*, 1974

[a] Radiation regime is presented as source, dose rate, and total dose; a dashed line indicates information was not available. Units are those used by the author.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

5.43 rad/h (4822 rad total dose). Mild hemopoietic atrophy in kidneys and spleens in some fish was seen after 128 d at 1.5 and 3.0 rad/h (4608 rad and 9216 rad total doses).

The immune response of chronically irradiated rainbow trout (*Salmo gairdnerii*) embryos has been examined by analyzing antibody synthesis against *Chondrococcus columnaris* disease in fish exposed to tritiated water (Strand *et al.*, 1973a). Developing embryos exposed to tritiated water for 20 d demonstrated a depressed immune response when compared to control fish.

Conclusions on Pathophysiology Data

With the exception of studies relating to immunity, research on the pathophysiological effects of radiation exposure in aquatic organisms has primarily served to elucidate that the mechanisms of radiation-induced mortality in aquatic organisms are similar to those observed in mammals. Thus, they are mainly studies on the cellular manifestations of lethal doses. Factors that modify these responses have not been widely studied (except for the effects of changes in salinity), but we propose that future work in this

Table 7. Induction of pathophysiological changes in fish from chronic exposure to radiation.

Endpoint	Organism/lifestage	Radiation regime[a]	Comments	Reference
Immune response: antibody synthesis against *Chondrococcus columnaris* disease	*Salmo gairdnerii*[b] (rainbow trout)/ embryos	Tritium 1 µCi/mL and 10 µCi/mL 20 d	Developing embryos exposed to tritiated water demonstrated depressed immune response	Strand *et al.*, 1973a
Hemopoiesis	*Gambusia affinis*[b] (mosquitofish)/adults	^{60}Co 0.5-5.43 rad/h 336-9216 rad	No demonstrable damage to hemopoietic tissues after 37 d at 5.43 rad/h (4822 rad); mild hemopoietic atrophy in kidney and spleen in some fish after 128 d at 1.0 and 3.0 rad/h (4608 rad and 9216 rad)	Cosgrove *et al.*, 1975

---

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time). Units are those used by the author.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

area would not be as productive as examining more sensitive endpoints such as reproductive and genotoxic effects and the factors that modify them. However, further studies on immunity in fishes may be warranted. In mammals, the pathophysiologic effect of primary concern at low doses is tumor induction in a variety of tissues. To our knowledge, only one study has reported data that demonstrate radiation-induced tumor production in aquatic organisms (Anders *et al.*, 1973a,b).

REPRODUCTION

Effects of ionizing radiation on fertility have been observed at relatively low dose rates and low total doses in mammals, fishes, and aquatic invertebrates. Our review focuses on these low-dose effects because other research utilizing relatively high doses has been reviewed previously (see Appendix I for references). Reproductive effects occur at lower doses than those causing mortality or typical pathophysiologic lesions, excluding tumor development in mammals. Effects of ionizing radiation on reproduction in mammals include responses at very low doses. For example, the LD_{50} of mouse oocytes has been reported to be 5 rad from tritium exposure and 8 R for x-ray exposure. The reported LD_{50} of monkey oocytes after tritium exposure is 8 rad. Complete sterilization was observed in female mice exposed 20 to 40 d postconception to 8.4 R/d from a ^{137}Cs source (in NRC, 1980). In discussing reproductive effects, it is important to note that in many studies in the aquatic literature, the induction of reproductive effects and dominant lethal mutations are not separated. For example, if an organism is irradiated and reductions in brood size are noted, they may be due to either gamete death or to dominant lethal mutations expressed in early development.

Reproductive Effects in Fish from Acute Radiation Exposure

Effects of ionizing radiation on reproduction in teleosts have been studied by several investigators (Table 8). However, Egami and co-workers performed more investigations in this area than any other group. For the purpose of this discussion, their work is divided into three categories. First, mechanistic studies were conducted that use high dose rates and total doses greater than 1000 rad (10 Gy). Because of the high doses used in these studies, which were reviewed in Egami and Ijiri (1979), they are not listed in

Table 8. Induction of reproductive changes in fish from exposure to acute levels of radiation.[a] Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[b]	Comments	References
Oncorhynchus tschawytscha[c] (chinook salmon)/embryos	x ray 250-1000 R	Counts of primordial germ cells in salmon irradiated at **250 R** were 10% of control values	Welander *et al*., 1948
Oryzias latipes[c] (medaka)/adult males	x ray 250-2000 rad	**500 rad** caused a significant decrease in oviposition frequency, an increase in percent of unfertilized eggs, and an increase in the number of sterile fish	Hyodo-Taguchi, 1980
Salmo gairdnerii[c] (rainbow trout)/ 29-d embryos	^{60}Co 33.5 R/min 0, 600, 800 R	Sterility was induced at lowest exposure tested (**600 R**) and at all observation times (60-150 d)	Konno, 1980

[a] Research on the reproductive effects of acute irradiation in teleosts has been reviewed by Egami and Ijiri, 1979. Because most of this research employed doses above 1000 rad (10 Gy), and has been reviewed extensively it is not listed above.

[b] Radiation regime is presented as source, dose rate, and total dose. For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[c] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

Table 8 and are not discussed. The second type of study conducted was the analysis of gamete death from acute doses below 1000 rad (10 Gy). These studies were also extensively reviewed in Egami and Ijiri (1979), but some are listed in Table 8 and are discussed in this section. The third type of study conducted has been the analysis of gamete death from doses delivered at relatively low dose rates. Results of this research are listed in Table 9 and are discussed in detail in the section on reproductive effects due to chronic radiation exposure.

Effects of acute ionizing radiation on female germ cells of teleosts have been observed after their exposures to doses as low as 250 R. Welander *et al.* (1948) found that counts of primordial germ cells in the chinook salmon *Oncorhynchus tschawytscha* exposed to 250 R from an x-ray source were 10% of control values. Konno (1980) exposed rainbow trout *Salmo gairdnerii* embryos to a ^{60}Co source (33.5 R/min) for total doses of 0, 600, and 800 R. Sterility, which was induced at the lowest dose tested, was detected at all observation periods (60 to 150 d).

Male germ cells of the medaka *Oryzias latipes* were studied extensively by Egami and co-workers, who found a temporary reduction in testicular weights after exposure to 100 to 2000 R (x-ray source). They also found that, in sexually inactive fish exposed to 100 to 1000 R, certain spermatogonial types and primary spermatocytes exhibited damage within 3 d (Egami and Ijiri, 1979).

Reproductive Effects in Invertebrates from Acute Radiation Exposure

Effects of acute irradiation on reproduction in aquatic invertebrates occur over a dose range of at least two orders of magnitude (Table 9). Causes for this broad range may not lie in actual species-specific differences in gamete killing, but in differences in the gamete stage irradiated and in the cell-repopulation capacity of different organisms.

In experiments by Ravera (1967), adults of the freshwater snail *Physa acuta* received 2000 to 220,000 R from an x-ray source. A dose of 2000 R reduced fertility and embryo viability. However, recovery from the damage to germ tissue was evident in snails receiving 2000 and 10,000 R. At 10,000 R, the production of egg capsules and the number of eggs per capsule were reduced. Moreover, all embryos produced during the first 50 d after irradiation from snails receiving 10,000 R were not viable. A greater percentage of the embryos

Table 9. Induction of reproductive changes in invertebrates from exposure to acute levels of radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Gammarus duebeni[b] (amphipod)/adults	x ray 65-740 R	Reduced egg-production rate at 220 R	Hoppenheit, 1973
Artemia salina[c] (brine shrimp)/adults	x ray 500 R and 1000 R	Progressive decrease in sensitivity of oocytes throughout prophase and into metaphase. Significant reduction in hatching after 500-R exposure	Cervini and Giavelli, 1965
Artemia salina[c] (brine shrimp)/adults	-- 1000-5000 R	Reduced fecundity at 1000 R, the lowest exposure tested	Squire, 1970
Physa acuta[b] (freshwater snail)/adults	x ray 2000-220,000 R	Reduced fecundity of the adults and reduced viability and fertility of the eggs was observed at 2000 R; temperature increase exacerbated radiation-induced effects	Ravera, 1967
Neanthes arenaceodentata[c] (polychaete)/adults and juveniles	^{137}Cs 500 rad/min 100-100,000 rad	No oocyte development was observed in adults receiving 50 Gy; reduced fecundity was observed between 1 and 5 Gy	Anderson *et al.*, in prep.
Physa acuta[b] (freshwater snail)/adults	x ray 2000-220,000 R	Reduced fecundity of adults; 100,000 R abolished reproductive capacity of adults	Ravera, 1966

---

[a] Radiation regime is presented as source, dose rate, and total dose; a dashed line indicates information was not available. For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Organism is exclusively marine.

produced after that time were viable. Ravera (1967) proposed that repopulation of the gonad by undamaged germ cells could explain this effect. Reproductive activity was completely abolished at 100,000 R.

Differential radiosensitivity between meiotic stages has been studied in prophase and metaphase oocytes of *Artemia salina*. Cervini and Giavelli (1965) showed that radiosensitivity of oocytes declined as prophase progressed. Metaphase oocytes were also less sensitive than prophase oocytes.

Hoppenheit (1973) observed reduced egg-production rate in adults of the amphipod *Gammarus duebeni* receiving as low as 220 R. However, this was offset by higher survival of adult females and increased brood size.

Anderson *et al*. (in preparation) found that brood size of the marine polychaete worms *Neanthes arenaceodentata* was reduced after doses between 1 and 5 Gy were delivered to adult worms. In contrast, preliminary data on irradiated juveniles indicated such effects occurred between 5 and 10 Gy, suggesting that repopulation of the gonad by undamaged germ cells may occur.

In determining the comparative sensitivity of gametes in selected species, it is important to know the gametogenic stage that was irradiated. However, this is not well characterized in some reports. Furthermore, both oogenesis and spermatogenesis are asynchronous in many aquatic organisms and, hence, many stages may be irradiated simultaneously.

Reproductive Effects in Fish and Invertebrates from Chronic Radiation Exposure

Chronic, low-level radiation studies may be conducted over many gametogenic stages and cycles. The lowest dose rate at which effects of chronic radiation exposure on fertility of aquatic invertebrates and fishes have been demonstrated (Tables 10 and 11) is between 0.59 R/d (Trabalka and Allen, 1977) and 10 R/d (Bonham and Donaldson, 1972). This is comparable to the range in which effects of chronic irradiation on fertility are first observed in mammals. These data indicate that fish and invertebrate oocytes are not more radioresistant than mammalian oocytes. However, when single acute doses of radiation are used to study reproduction, lower organisms may have a higher capacity to replace damaged cells.

At least five studies on fishes and aquatic invertebrates were conducted in which the effect of chronic radiation exposure in the dose-rate range mentioned above were examined (Tables 10 and 11). Trabalka and Allen (1977) compared populations of the mosquitofish *Gambusia affinis* from the

Table 10. Induction of reproductive changes in fish from chronic exposure to radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Gambusia affinis[b] (mosquitofish)/all stages	White Oak Lake 0.59 R/d lifetime	No decrease in fecundity observed at 0.59 R/d; however, increased embryo mortality was noted.	Trabalka and Allen, 1977
Poecilia reticulata[b] (tropical guppy)/ 0-3 d embryos	^{137}Cs 4.08, 9.60, 30.48 rad/d Total number of days varied	Total fecundity was markedly reduced at all dose rates because of a decrease in mean actual brood-size and an increase in temporary and permanent infertility; the lowest dose rate tested was **4.08 rad/d**	Woodhead, 1977
Poecilia reticulata[b] (tropical guppy)/ 0-3 d embryos	^{137}Cs 6.0, 12.0, 40.8 rad/d Total number of days varied	One pair out of 6 and 5 out of 7 were sterile at 6.0 and 12.0 rad/d, respectively; total accumulated doses were 4000 and 8000 rad; at 40.8 rad/d complete sterility occurred at 5000 rad; this marked dose-rate effect was not reflected in changes in fecundity in fertile guppies	Purdom and Woodhead, 1973
Oryzias latipes[b] (medaka)/adult males	^{137}Cs 1.3-84.3 R/d 78-5058 R	408 R delivered at 6.8 R/d caused a decreased oviposition frequency, an increased percent of unfertilized eggs, and an increase in number of infertile fish	Hyodo-Taguchi, 1980
Oncorhynchus tschawytscha[b] (chinook salmon)/smolts	^{60}Co 0.5-50 R/d 40-4000 R	Gonadal development was retarded at 10 R/d or more	Bonham and Donaldson, 1972

---

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time). Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

Table 11. Induction of reproductive changes in invertebrates from chronic exposure to radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Physa heterostropha[b] (aquatic snail)/adults	White Oak Lake 0.65 rad/d lifetime	Frequency of egg-capsule production was reduced in the irradiated (**0.65 rad/d**) population, but an increase in number of eggs per capsule occurred, resulting in similar rates of egg production compared to controls	Cooley, 1973
Physa heterostropha[b] (aquatic snail)/adults	^{60}Co 24, 240, 600 rad/d 24 weeks	Egg capsule and egg production were completely stopped by 600 rad/d, were reduced significantly by 4 weeks and stopped completely.by 19 weeks at 240 rad/d, and were only significantly lower than controls during week 4 to 12 for the group receiving **24 rad/d**	Cooley and Miller, 1971

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time). Units are those used by the author.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

radionuclide-contaminated White Oak Lake at the Oak Ridge National Laboratory to those from a matched control site. They found no decrease in fecundity, but an increase in embryo mortality of the fish from White Oak Lake. These results were obtained by returning animals collected from radionuclide-contaminated and control sites to the laboratory and observing subsequent reproduction under controlled conditions. This study is more definitive than others conducted on animals from White Oak Lake, but results are confounded by the fact that contaminants other than radionuclides are present in White Oak Lake. Cooley (1973) examined the reproductive biology of pond snails at White Oak Lake. He also transplanted animals from White Oak Lake to the laboratory. He found that frequency of egg-capsule production was reduced; however, an increased number of eggs per capsule was also documented. It is interesting to note that a prior laboratory study by Cooley and Miller (1971) documented clearcut reproductive decline at 240 rad/d but not at 24 rad/d. Irradiation was initiated on 45-d-old snails, and laboratory effects might have occurred at lower levels if irradiation was extended over the entire lifetime of the organism.

Three of the five studies mentioned were conducted exclusively in the laboratory, where control of confounding variables is more readily achieved. The most rigorous of these studies was that of Woodhead (1977), who examined fecundity of the guppy *Poecilia reticulata* receiving 4.08, 9.60, and 30.48 rad/d. Total fecundity was significantly reduced at all dose rates. Reductions in fecundity in this important and thorough study are probably due both to reproductive effects (damage to gametes) and the induction of dominant lethal mutations in gametes. In a similar study, Woodhead *et al*. (1983) reported that *Ameca splendens* is even more sensitive than the guppy. However, this report is an abstract, and the details of the research cannot be evaluated at this time. Hyodo-Taguchi (1980) observed an increased percentage of unfertilized *Oryzias latipes* eggs after males used to inseminate the eggs received 6.8 rad/d for 60 d. No statistically significant effects were observed at 2.9 rad/d, the next lower dose rate used. Bonham and Donaldson (1972) exposed chinook salmon *Oncorhynchus tschawytscha* embryos for the first 80 d of life to 0.5 to 50 R/d. Approximately 4 weeks after the irradiations were completed, gonadal development was observed in smolts. They found that gonadal development was first retarded in those receiving 10 or more R/d.

Conclusions on Reproduction Data

Comparisons of the effects of acute irradiation on reproduction of fish and invertebrates are often not valid because the gamete stage irradiated differed between studies. Furthermore, even when the same stage was irradiated, it was not established that the capacity for repopulation of damaged cells was the same between organisms. Research on effects of chronic irradiation on the gonads is of particular interest, however, because the results show effect levels comparable to those observed in mammals. Dose rates between 0.5 R/d to 10 R/d appear to define a critical range in which detrimental effects on fertility are first observed in a variety of sensitive organisms. Unfortunately, almost nothing has been done on this subject with invertebrates. For aquatic organisms, nothing is known about whether responses would occur at lower levels if effects of modifying environmental factors were examined. The only definitive study on the effects of lifetime irradiation on the reproduction of aquatic organisms is that of Woodhead (1977). He demonstrated decreased fecundity in the guppy at 4.08 rad/d. Finally, it must be re-emphasized that the dose rates mentioned above are slightly higher than the highest dose rates observed in the ocean due to waste-disposal activities (0.240 rad/d) and considerably higher than those expected at deep ocean disposal sites.

DEVELOPMENT

Effects of radiation on the development of fish embryos has been the most widely debated topic in aquatic radiobiology (Woodhead, 1984), and results on irradiation of developing embryos by introduction of radionuclides directly into the test water have been most contentious. Recently, this topic was exhaustively reviewed by Woodhead (1984). Our purpose is to summarize some of the general conclusions of Woodhead (1984) and to identify the studies demonstrating effects at relatively low levels from (1) acute irradiation, (2) chronic irradiation from a sealed source, and (3) chronic irradiation by radionuclides in the water. Data on the effects of acute and chronic irradiation of aquatic invertebrate embryos are also reviewed. Tables 12-15 contain representative data on these subjects. No data on studies employing radionuclides in the water, other than tritium, are included.

Developmental Effects from Acute and Chronic Irradiation of Fish

Careful dosimetry is important in all radiobiological experiments, particularly so when radionuclides are present in the water rather than in a sealed, external source or generated by an x-ray machine. Accumulation of radionuclides in embryos and absorption of radionuclides onto membranes may affect the dose received by an organism. Woodhead (1984) concluded that all studies on fish embryos using radionuclides in the water (except those using tritium because it is not bioconcentrated) contain minor to very serious flaws in dosimetry. This means that, for such experiments, the dose received by the organism cannot be characterized. Because of this criticism, results from such studies are presented separately.

Different criteria have been used to evaluate the effects of ionizing radiation on developing fish embryos (Table 12): hatching success, embryo mortality, and frequency of morphologically abnormal embryos and larvae. The forms of the gross abnormalities in fishes include anophthalmia, cyclopia, monophthalmia, microphthalmia, abdominal and caudal foreshortening, abdominal lordosis, scoliosis, and abnormal fin structure. The relatively minor changes in fishes include changes in standard length, eye diameter, head length, pigmentation, and fin-ray number. From the differences in sensitivities at successive early stages of development, it appears that a number of different vulnerable processes must be involved in the production of maldevelopment and that these processes are in progress at various times considerably prior to the earliest histological evidence of organogenesis (McGregor and Newcombe, 1968).

Several experiments have demonstrated a trend of decreasing radiosensitivity with increasing development (Woodhead, 1984). For example, Bonham and Welander (1963) determined the LD_{50} at hatching and at 150 d after fertilization for the silver salmon irradiated at the one-cell stage. The minimum values were 30 R and 16 R, respectively, for fish irradiated at the one-cell stage. A consistent decrease in radiosensitivity of the embryos with increased age at the time of irradiation was observed in the later stages with an LD_{50} of 1871 R at hatching for irradiation in the late-eyed stage. These differences in radiosensitivity are related to critical periods in organogenesis and changes in mitotic activity.

Allen and Mulkay (1960) investigated the histological effects of exposure of the paradise fish Macropodus opercularis to 1000 R at different stages of development. The order of decreasing sensitivity of tissue and organ systems

Table 12. Induction of developmental changes in fish from exposure to acute levels of radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Oncorhynchus kisutch[b] (silver salmon)/one-cell embryos	x ray 12-2400 R	LD_{50} at **16 R** observed 150 d after fertilization	Bonham and Welander, 1963
Salmo gardnerii[b] (rainbow trout)/embryos	x ray 25-2570 R	Retardation in growth seen, at exposures as low as **38 R** for those irradiated during early-eyed stages; $\geq$ 200 R required for changes in other stages; increased frequency of abnormalities in irradiated embryos	Welander, 1954
Pleuronectes platessa[c] (plaice)/embryos	x ray 30-150 rad	LD_{50} at metamorphosis for plaice larvae irradiated at blastula stage was **90 rad**	Ward *et al*., 1971
Fundulus heteroclitus[c] (mummichog)/embryos	x ray --	Eggs at **300-400 R** in the one-cell to two-cell stage developed major malformations; fertilized eggs and early cleavage stages showed greatest sensitivities	Rugh and Clugston, 1955
Oncorhynchus tschawytscha[b] (chinook salmon)/embryos	x ray 250-10,000 R	Development of the eye, gill epithelium, fin, and chromatophores were studied; slight differences in pigmentation and retardation of gill-filament development were noted at **500 R**; otherwise, no effects were observed below 1000 R	Welander and Donaldson, 1948
Cyprinus carpio[b] (carp)/one-cell embryos	^{60}Co 7.1 rad/sec 50-2000 rad	LD_{50} at hatching was 601 rad for β radiation and **501 rad** for γ radiation; no hatching at 1000 rad	Blaylock and Griffith, 1971

Table 12. (Continued.)

Organism/lifestage	Radiation regime[a]	Comments	References
Salmo gairdnerii[b] (rainbow trout)/embryos	x ray 10, 100, and 1000 rad	No significant increase in malformations and mortality at 10 and 100 rads; significant increases at **1000 rad**; malformations the same as those from irradiating gametes	McGregor and Newcombe, 1968
Oncorhynchus tschawytscha[b] (chinook salmon)/embryos	x ray 250-10,000 R	Lowest exposure at which significantly decreased weight and length were observed was **1000 R**	Welander and Donaldson, 1948
Oryzias latipes[b] (medaka)/fertilized ovum and sperm through last embryonic stage	^{137}Cs 250, 33.33, and 1.7 R/min 2000 R	Decreased % hatch and increased incubation time from **2000 R** delivered at 250 R/min and 33.3 R/min, none at 1.7 R/min; lowering the temperature during irradiation negated dose-rate effects.	Egami and Hama, 1975
Oryzias latipes[b] (medaka)/fertilized ovum and sperm through last embryonic stage	x ray 2000 R	Changes in the hatch rate from a exposure to **2000 R** depending on what stage irradiated (pattern irregular)	Egami and Hama, 1975
Pleuronectes platessa[c] (plaice)/embryos	x ray 25-4800 R	No significant reduction in % hatching, malformations, and mortality	Templeton, 1966

---

[a] Radiation regime is presented as source, dose rate, and total dose; a dashed line indicates information was not available. For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Organism is exclusively marine.

was the following: blood and hemopoietic tissue > eye > central nervous system > germ cells > muscles > gut > heart > ear and lateral line > pronephros > olfactory organ > notochord > pigment cells. For all tissues and organs, it was found that specific types of damage to a presumptive tissue could be elicited by radiation prior to the appearance of its identifiable precursor cells. Also, in agreement with the law of Bergonie and Tribondeau, the period of differentiation of a tissue and any period of increased mitotic activity in a tissue or organ were found to result in increased radiosensitivity (Woodhead, 1984).

These data on organismal and tissue radiosensitivity in fish embryos raise two more important points relevant to our analysis of low-effect levels. First, the embryo or larval stage at which the radiation dose is received is a major determinant of what the effect level will be. Second, the time period over which observations are made must be sufficiently long for the induced effects to be observable.

The lowest effect levels observed from acute irradiation of developing fish embryos are the levels observed by Bonham and Welander (1963) described above (Table 12). Thus, fish-embryo mortality has been documented from exposures of 16 R. This low value is lower than minimum-effect levels for other species. Blaylock and Griffith (1971) found the LD_{50} at hatching for carp embryos exposed as single-cell eggs to be 601 rad for β radiation and 501 rad for γ radiation. An LD_{50} of 90 rad at metamorphosis was determined for plaice larvae, the only exclusively marine fish studied, irradiated at blastula stage (Ward et al., 1971).

Very few studies have been conducted that use sealed sources to examine the effects of chronic irradiation on fish embryos (Table 13). As a part of a larger study on the effects of chronic irradiation on chinook salmon embryos, Donaldson and Bonham (1964) found a significant increase in opercular defects of smolt at exposures of 33 to 49 R given at 0.5 R/d from the moment of fertilization.

In contrast to the paucity of studies using external sources to examine the effects of chronic irradiation on fish embryos, numerous studies exist in which radionuclides have been introduced directly into the test water. However, Woodhead (1984) has concluded that these studies have contributed

Table 13. Induction of developmental changes in fish from chronic exposure to radiation.[a] Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[b]	Comments	References
Oncorhynchus tschawytscha[c] (chinook salmon)/embryos *Oncorhynchus kisutch*[c] (coho salmon)/embryos	^{60}Co 0.5 R/d 33-40 R	Significant increase in opercular defects in embryos given 0.5 R/d when compared to controls	Donaldson and Bonham, 1964
Oncorhynchus tschawytscha[c] (chinook salmon)/embryos *Oncorhynchus kisutch*[c] (coho salmon)/embryos	^{60}Co 0.5 R/d 33-40 R	In some phases, growth was superior in the irradiated (0.5 R/d) group with no discernable pattern; a greater average weight in the irradiated group was statistically significant	Donaldson and Bonham, 1964
Poecilia reticulata[c] (guppy)/embryos and young	Tritium 0.05 to 1 mCi/mL 340-4700 rad for young fish and 90-2500 rad for embryos	Total doses of 3400 and 4700 rad for young fish and of 380 rad for embryos resulted in significantly lower weights; significant increases in weight seen in males receiving 2500 rad as embryos; increased proportion of males and earlier appearance of sex characteristics in males receiving 90-rad total dose during embryogenesis	Erickson, 1973
Salmo gairdnerii[c] (rainbow trout)/embryos	Tritium 0.01, 0.1, 1.0, and 10 µCi/mL 25 d	No significant decrease in hatching; slight enhancement in hatching at 0.01 and 0.1 µCi/mL; no significant increase in malformations	Strand *et al.*, 1973b
Gasterosteus aculeatus[c] (stickleback)/embryos	Tritium 0.5, 1.0, and 2.0 mCi/mL 980-3920 rad	Significant reduction in mean eye diameter in 1.0- and 2.0-mCi/mL water	Walden, 1973

---

[a] Most studies on developing fish embryos directly exposed to radionuclides in the test water are not reported herein. They have been critically reviewed by Woodhead (1984) and are discussed briefly in the text.

[b] Radiation regime is presented as source, dose rate, and total dose (or exposure time). Units are those used by the author; R is the abbreviation for roentgen.

[c] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

little to the field of aquatic radiobiology because of the dosimetry problems described above. With regard to determination of low-effect levels, Woodhead (1984) concluded the following:

> Using the criterion of embryo survival (as measured by hatching rate) after irradiation for the major part of embryogenesis, the highest total absorbed dose at which no effect has been observed is 8600 rad (Till, 1976, 1978), while the lowest at which an effect has been noted is 0.3 rad (Strand and co-authors, 1973b). This discrepancy may well be much greater, since the former exposure was derived from ^{238}Pu while the latter was due to tritium, and the relative biological effectiveness of the α radiation would be expected to be greater than that of the β radiation. There are 2 points which generate some reservations about the validity of the lower limit. Strand and co-authors (1973b) in fact report 2 experiments in which the hatching success was measured, and 2 concentrations of tritium are duplicated between these experiments. In the first experiment no response was noted in respect of (sic) hatching success at these two concentrations, whereas in the second, such an effect was observed. Thus there can be some doubt as to whether a genuine radiation-induced effect was being produced. This conclusion is supported by the fact that the decline in hatching success was not found to be monotonically dependent on increasing dose rate; the highest dose produced less effect on hatching success than either of the 2 lower doses. If these data are excluded on the basis of these apparent internal inconsistencies, then the remaining doses at which an effect has been observed are 2160 rad (Ichikawa and Suyama, 1974) and 750 rad (Fedorova, 1972b). Since Fedorova (1972b) only gives data for a single concentration of ^{14}C, it is not possible to be certain that 750 rad represents the lower limit for a significant effect on the hatching success of *Coregonus peled*. From the data given in the paper it can also be concluded that the actual dose to the embryo could be as much as 50% greater, i.e. 1130 rad. Fedorova's data also provide confirmation that a significant part of the total mortality is caused by a small fraction of the total dose delivered in the early part of embryogenesis, and also that the embryo becomes less sensitive during the course of development.
>
> There are only 2 sets of data on the incidence of embryo abnormality (Strand and co-authors, 1973b; Till, 1976, 1978). Once again, the data of Strand and co-authors, (1973b) can only be accepted with reservations due to the clear lack of consistency within and between the 2 experiments reported; the embryos receiving the highest absorbed dose yielded a lower proportion of anomalous individuals than any other treatment, including the 2 control populations. Till (1976, 1978) observed that the lowest dose at which a significant increased proportion of abnormal embryos was produced was 3400 rad.

Woodhead (1984) also delineated the following improvements necessary for future studies using radionuclides directly in the test water:

1. Full details must be given of the differential accumulation of the radionuclide(s) by the eggs and the components of their environment, as a function of time at each radionuclide concentration.

2. Estimates of the absorbed dose to the embryo and its variation during development must be made, and full details of the derivation given.

3. Sufficient replicates must be made to permit the relative statistical significance of any observation to be determined.

4. A range of radionuclide concentrations must be employed so as to allow the construction of a reliable dose-effect curve over the range of 0 to 100% response. Experiments performed with this degree of attention to detail will not be easy or straightforward.

Almost nothing is known about the effect of modifying environmental factors on the response of fish embryos to radiation. However, the modifying effects of dose rate and temperature on response of fish embryos to irradiation were examined by Egami and Hama (1975). They found that 2000-R exposures at early stages of development reduced hatching rate and increased development time when the exposure rate was 250 R/min or 33.3 R/min, but not at 1.7 R/min. Decreasing the temperature of the eggs during the exposure period negated these dose-rate effects.

Developmental Effects from Acute and Chronic Irradiation of Invertebrates

Few studies exist that chronicle the effects of radiation on development of invertebrate embryos (Tables 14 and 15). Blaylock and Trabalka (1978) cite studies using radionuclides in the water. These will not be reviewed here because the doses received by the test organisms were not documented adequately. Studies by Ravera on Physa acuta (1968), cited in Blaylock and Trabalka (1978), are described in the section on mortality. Similar to results on fish, an increasing radiosensitivity with age was noted for this snail. The

Table 14. Induction of developmental changes in invertebrates from exposure to acute levels of radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Diaptomus clavipes[b] (calanoid copepod)/adults and embryos	^{60}Co 7000 rad/min 1000-100,000 rad	Significant decrease in % hatch at lowest dose, **1000 rad**	Gehrs *et al.*, 1975
Physa acuta[b] (freshwater snail)/embryos	x ray 200-10,000 R	Decrease in percent hatching seen at exposures as low as **2000 R**	Ravera, 1966
Physa acuta[b] (freshwater snail)/embryos	--	LD_{50} values for the four-cell, trochophore, veliger, and hippostage embryos of the gastropod *Physa acuta* were **1075**, 2350, 4900, and 10,750 R, respectively	Ravera, 1968 (in Blaylock and Trabalka, 1978)
Artemia salina[c] (brine shrimp)/cysts	^{60}Co 12,000 rad/min 60,000-960,000 rad	Approximately 25% decrease in hatchability of irradiated cysts at **360,000 rad**	Iwasaki, 1965

[a] Radiation regime is presented as source, dose rate, and total dose; a dashed line indicates information was not available. For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Organism is exclusively marine.

Table 15. Induction of developmental changes in invertebrates from chronic exposure to radiation.

Organism/lifestage	Radiation regime[a]	Comments	References
Physa heterostropha[b] (aquatic snail)/ adults and embryos	^{60}Co 1, 10, 25 rad/h 25 weeks	Significant decrease in % hatching at 10 rad/h but not at 1 rad/h; no hatching at 25 rad/h; growth of adults was significantly increased by 10 and 25 rad/h	Cooley and Miller, 1971

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time). Units are those used by the author.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

lowest observed effect level was an LD_{50} of 1075 R for embryos with four cell stages. Gehrs *et al.* (1975) administered 1000 to 100,000 rads to embryos of the freshwater copepod *Diaptomus clavipes*. A significant decrease in the hatching rate was observed at the lowest dose.

Conclusions on Development Data

Mortality of developing fish embryos from acute irradiation has been observed at as low a dose as 16 R (Bonham and Welander, 1963; see mortality section for further comments). Increased developmental abnormalities were documented by Donaldson and Bonham (1964) after exposures of 0.5 R/d. They initiated these 80-d experiments with one-cell chinook salmon embryos. Similarly, developmental effects have been observed in mammals at dose rates as low as 2 to 3 R/d. In NRC (1980) are cited studies indicating increased prenatal and postnatal mortality from exposures of 2.5 R/d in rats irradiated from conception to term.

Woodhead (1984) concluded that, with maximum dose rates (related to waste disposal) of 0.240 rad/d observed in the ocean, fish embryos would not be at increased risk. However, he also believed a final uncertainty remained, with the lack of convincing work on the introduction of radionuclides directly into the test water. We accept these conclusions, but also feel that additional research on the effects of modifying environmental factors is necessary to determine the error associated with our current knowledge of low observed effect levels. Detrimental effects in fish have been demonstrated at 0.5 R/d, a value only twice the highest dose rates observed in the ocean. Considering the findings (Egami and Hama, 1975) that low temperature may negate dose-rate effects, it is conceivable that effect levels at low temperatures may be lower than those discussed above.

Finally, we conclude that there is not sufficient information on chronic or acute irradiation of developing embryos of aquatic invertebrates. More work is needed in this area.

GENETICS

The genetic effects of ionizing radiation have been a principal concern of researchers in mammalian radiobiology. Genetic effects are particularly important because some effects are known to occur at low dose rates (NRC, 1980; UNSCEAR, 1982; UNSCEAR, 1977). The major somatic and heritable genetic effects of low-level radiation have been identified for mammalian populations (NRC, 1980; UNSCEAR, 1982; UNSCEAR, 1977). If one accepts the theory that mutational events in somatic cells are primarily responsible for cellular transformation and tumor formation, then the principal genetic effect of low-level radiation in somatic tissue is cancer induction. Concern over effects on germ cells of low-level radiation centers on inducing increased frequency of heritable genetic disease. This may occur from mutation, chromosome breakage, chromosome rearrangement, or faulty segregation of chromosomes at metaphase which may lead to aneuploidy. The significance of increased frequency of neutral mutations is also the subject of considerable debate (Newcombe, 1971).

Gene mutations may have negative effects (cell death or genetic disease), positive effects (increased population heterozygosity), or neutral effects (those that cannot be characterized). Although evidence exists for increased population fitness due to positive or neutral mutations, most experts concur that increases in mutation rates should be considered detrimental (Newcombe, 1971; UNSCEAR, 1977;).

Impact of increased frequency of heritable genetic diseases (due to gene mutations, chromosomal damage, or aneuploidy) and their potential for individual suffering has been of greater concern to those interested in human-health effects than to those interested in environmental effects. However, genetic effects on populations are of concern to those interested in both human and environmental health. Many scientists predict that any mutation causing deleterious effects in populations would rapidly be eliminated by natural selection. This may be true, but it should be remembered that such a statement does not presume no effect at the organismal level.

Dominant lethal mutations also occur as a result of radiation. These are chromosomal or gene mutations that result from irradiation of developing sperm or eggs and cause early embryo death. Such mutations have consequences at the population level if a high percentage of embryos from a spawn are affected. Viability of embryos produced from irradiated adult fish is an index of dominant lethal mutations and is referred to earlier in this report.

Production of chromosomal aberrations, which may also result in cell death, is a well-documented effect of radiation and is the result of breakage with or without exchange of chromosomal material. When stable aberrations such as translocations occur in germ cells, they may be transmitted to subsequent generations. Metaphase analysis of chromosomal aberrations in human lymphocytes has been used for biological dosimetry on human populations.

Because the genetic effects of ionizing radiation are of central importance in mammalian radiobiology, several investigators working with aquatic organisms have realized the need for genetic-effects research on aquatic organisms (Kligerman, 1980; Metalli, 1979; Harrison *et al.*, 1985). The status of this research to date is summarized in the following sections.

General Considerations in Aquatic Genotoxicity Research

Selection of endpoints, model species, and other basic aspects of study design are important general considerations in aquatic genotoxicity research. To date, both mutations at a specific genetic locus and dominant lethal mutations have been studied in aquatic organisms, and the induction of chromosomal aberrations has been studied by metaphase chromosome analysis.

Studies on the induction of specific locus mutations are limited by lack of knowledge about the genetics of most aquatic organisms, as well as the long time required and the great expense of conducting this type of experiment (Kligerman, 1980). However, measuring dominant lethal mutations is relatively easily achieved in a variety of fish and invertebrates in which gametes and developing embryos may be readily obtained from irradiated adults.

A variety of cytogenetic endpoints are available to examine the effects of radiation on aquatic organisms. These are summarized in Fig. 2. These endpoints include analysis of chromosomal aberrations in metaphase and in interphase by premature chromosome condensation, analysis of sister chromatid exchange (SCE), the micronucleus assay, and the anaphase aberration assay. Metaphase analysis of chromosomal aberrations is the most laborious, but it is also the most highly recommended endpoint (Kligerman, 1979; Metalli, 1979).

Selection of species for cytogenetic studies has been discussed by Kligerman (1979). He lists five criteria for a model organism for _in vivo_ cytogenetics:

1. A satisfactory karyotype
2. Tissues that yield adequate numbers of well-spread metaphases
3. Ability to withstand experimental conditions
4. Ease of obtaining and maintaining in the laboratory
5. Relatively small size

To this list, we must add a few more criteria. First, the cell kinetics of the tissue to be studied should be known or be characterized in advance. This is because at each cell division, certain percentages of aberrations or micronuclei are lost from the cell. This should be taken into account when species or tissues are being considered for analysis. Because tissue kinetics are often complex, it is also recommended that a single cell type within an organism, rather than a mixed tissue, be used whenever possible. Finally, it is preferable to select a tissue that does not have a restricted seasonal availability. An example of such a tissue would be oogonial or spermatogonial tissue in fish and invertebrates with highly seasonal reproductive cycles. Satisfaction of each of these criteria in any one study may not be possible at this time; however, these are important guidelines for future research.

Cytogenetic Endpoint	Types of Damage	Advantages	Disadvantages
Analysis of chromosomal aberrations	Breakage or breakage and exchange of chromosomes	● Very sensitive ● Types of aberrations induced can be identified	● Tedious ● Many cells required for analysis
Sister chromatid exchange (SCE)	Exchange of chromosomal material between sister chromatids; significance is unknown, but SCE has been correlated with increased mutation frequency	● Very sensitive ● Highly quantitative ● Fewer cells required than for metaphase aberration analysis	● 1 or 2 cell divisions must occur in thymidine analogue; this limits some field applications ● Biological meaning is still uncertain ● More sensitive to chemical damage than radiation damage
Micronucleus assay	Chromosomal breakage and spindle malfunction causing laggard chromosomes	● Sensitive ● Quantitative ● Relatively simple preparation and analysis	● Scoring may be complicated in mixed tissues or cells with large nuclei ● Single-cell suspensions are required ● Knowledge of cell kinetics is essential
Anaphase aberration assay	Chromosomal breakage, translocations, spindle malfunction, and chromosome stickiness	● Relatively simple preparation and analysis	● Because anaphase figures cannot be accumulated, use of this assay is restricted to rapidly dividing tissue such as embryos ● Different orientations of anaphase figures may give different results

Figure 2. Important endpoints in cytogenetic research and their implications.

Induction of Chromosomal Aberrations in Fish

Three studies were performed in which chromosomal aberration induction in fish *in vivo* was examined (Table 16). In the first study, Kligerman *et al.* (1975) introduced the fish *Umbra limi* as a model for the study of chromosomal aberrations in fishes. This animal has a favorable karyotype of 22 large metacentric and submetacentric chromosomes, and sufficient numbers of metaphases from a variety of tissues are obtainable for analysis. They administered a single exposure of radiation (325 R, x rays) to a group of fish and compared aberration rates in this group to those in a control group. The rate in the control was 0.03% aberrant metaphases per fish; in the experimental group, it was 30% aberrant metaphases per fish.

Mong and Berra (1979) conducted a similar study on *Umbra limi* using exposures of 350 R, 660 R, and 990 R from an x-ray source. They detected 9.2%, 13.8%, and 20.5% aberrant metaphases, respectively. In these two studies, little attention was paid to important methodological details. For example, in neither study was a clear breakdown of the types of aberrations observed presented nor was it required that the entire chromosome complement be visible, or that scored the cells be scored blind. Furthermore, mixed tissues were analyzed without any discussion of cell kinetics or mitotic delay.

In the third *in vivo* study, Suyama *et al.* (1980) examined anaphase translocation bridges induced by tritium exposure in embryos of the plaice *Limanda yokohamae*. A significant increase over control values was detected in the group exposed to 1 Ci/L for 22 h, but not at 0.1 Ci/L.

Two important *in vitro* studies (Table 16) were conducted on fish cells, and significant effects were observed at the lowest radiations tested (50 R and 75 rad). In the first study, Woodhead (1976) cultured *Ameca splendens* embryonic tissue and delivered 75 to 1070 rad to these cells. In an attempt to deal with cell kinetics, mitotic delay, and differential sensitivity at different stages in the cell cycle, he sampled at 21, 45, and 69 h. He used most of the characteristics of good cytogenetic study, such as blind scoring and rigorous scoring criteria, in this work. In Fig. 3 is a summary of the data and a comparison with other species. It is evident that the response obtained was similar, but not identical, to that obtained by other investigators for the toad *Bufo marinus* and human *Homo sapiens* cells.

Table 16. Induction of chromosomal aberrations in fish from acute and chronic exposure to radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Umbra limi[b] (central mudminnow)/ lymphocytes, *in vitro*	x ray 50-200 R	Cultured lymphocytes were irradiated and examined for the presence of dicentrics only; at the lowest exposure, **50 R**, 16 dicentrics were observed in 424 cells (0.038 dicentrics per cell); whereas none were observed in 571 control cells	Suyama and Etoh, 1983
Ameca splendens[b] (--)/ embryonic tissue, *in vitro*	^{60}Co 560 ± 20 rad/min 75-1070 rad	Aberrations significantly above control levels were detected at the lowest dose, **75 rad**, using embryonic tissue *in vitro*; response level was comparable in sensitivity to that of other vertebrates	Woodhead, 1976
Umbra limi[b] (central mudminnow)/adults	x ray 325 R	Chromosomal aberrations were observed in approximately 30% of the metaphases examined per fish after an exposure of **325 R**; the control rate was 0.03%	Kligerman *et al*., 1975
Umbra limi[b] (central mudminnow)/adults	x ray 350-990 R	9.2%, 13.8% and 20.5 % aberrant metaphases were detected at **350 R**, 660 R, and 990 R, respectively	Mong and Berra, 1979
Limanda yokohamae[c] (plaice)/embryos	Tritium 0.1, 1, 10 mCi/mL 22 h	Plaice embryos irradiated with 1 and 10 mCi/mL immediately after fertilization showed significantly more mitoses with translocation bridges than did controls	Suyama *et al*., 1980

---

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time). For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Organism is exclusively marine.

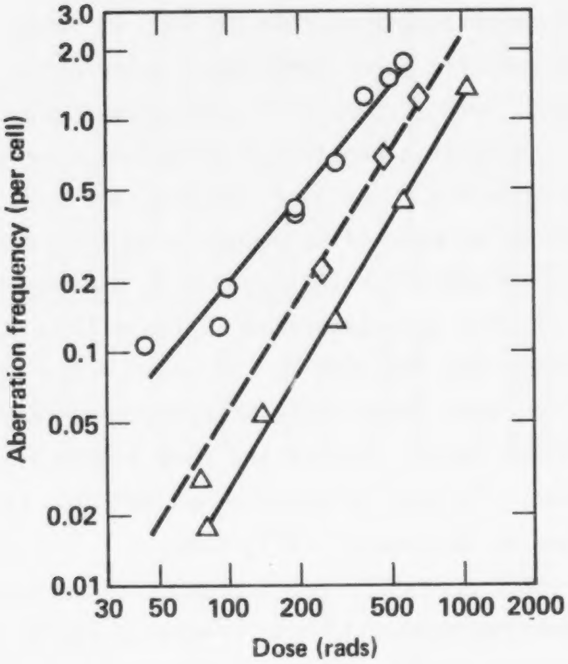


Figure 3. Comparison of yields of dicentrics and centric rings in fish (A. splendens, Δ), toad (B. marinus, ◇), and man (H. sapiens ○) (Woodhead, 1976).

In the second *in vitro* study, Suyama and Etoh (1983) examined x-ray-induced dicentric yields in cultured lymphocytes of *Umbra limi*. The dicentric yields were significantly increased at 50 R, the lowest exposure tested. However, only dicentrics were scored, and they were sometimes scored from metaphase spreads in which only 20 or 21 of the 22 chromosomes were visible. It is not clear whether or not the slides were coded for blind scoring.

Induction of Mutations in Fish

Induction of specific locus mutations and dominant lethal mutations have been examined in a variety of studies on fish. However, in most of these studies, relatively high exposures (500 to 1000 R) were used.

In studies on specific locus mutations, color-pattern mutation rates (Purdom and Woodhead, 1973), changes in aggressive behavior (Holzberg and Schroeder, 1972), and genetic regulation of melanophore genes (Anders *et al*., 1973a,b) have been examined. The last study is the most notable because it also is the only study we know of to demonstrate the induction of premelanomas and melanomas in irradiated fish (*Platypoecilus maculatus* and *Platypoecilus variatus*). These studies are summarized in Table 17. Effects were observed from exposures between 500 and 1000 R. In view of the time required and expense of conducting such large-scale breeding experiments, cytogenetic endpoints and dominant lethal studies are more highly recommended for research on aquatic organisms. Further information on mutation research in teleosts is available in reviews by Schroeder (1979, 1980).

Rates of embryonic mortality and incidence of severe embryonic abnormalities in embryos produced from irradiated adults or irradiated gametes are indices of dominant lethal mutations. These are to be distinguished from observations of these same effects in organisms irradiated as embryos; effects from irradiation of embryos are indicative of developmental abnormalities.

Table 17 summarizes the many studies in which dominant lethal mutations are examined (some studies in the section on reproduction have also involved the analysis of dominant lethal mutations.) However, all except one used relatively high exposures. The study that demonstrates the lowest effect level is that of McGregor and Newcombe (1972a). They administered doses of 25 to 400 rad to rainbow trout *Salmo gairdnerii* sperm and examined the induction of major eye malformations in F_1 progeny. A significant increase was observed at the lowest dose.

Table 17. Induction of mutations in fish from acute and chronic exposure to radiation. Entries are ordered according to the lowest dose or dose rate at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime^a	Comments	References
Gambusia affinis^b (mosquitofish)/all stages	White Oak Lake 0.59 rad/d lifetime	A significant increase in frequency of dead embryos in the irradiated (0.59 rad/d) population versus control was demonstrated; this indicated the presence of radiation-induced recessive lethal mutations maintained in the population since initial studies in 1966	Trabalka and Allen, 1977
Salmo gairdnerii^b (rainbow trout)/sperm	^{60}Co 64 rad/min 25-400 rad	Major eye malformations were induced in F_1 progeny when sperm received 25 rad, the lowest dose tested; frequencies were linear between 25 and 400 rad, with a doubling dose of 54 rad; in contrast, decreased embryo mortality was observed at 25 and 50 rad	McGregor and Newcombe, 1972a,b
Salmo gairdnerii^b (rainbow trout)/gametes	x ray 200-20,000 rad	Major malformations from exposure of eggs or sperm; at 200 rad, the yield of malformations was 300 per million embryos per rad; malformations appear to be associated with gross chromosomal changes	Newcombe and McGregor, 1967
Poecilia reticulata^b (tropical guppy)/adults	x ray 500 and 1000 R	Vertebral column abnormalities were observed in F_1 progeny of adults given 500 and 1000 R while germ cells were immature	Schroeder, 1969a
Platypoecilus maculatus^b and *Platypoecilus variatus*^{b} (platyfish)/adults	x ray 500-1500 R	Premelanomas and melanomas were induced at 500-1500 R in studies on the genetic regulation of melanophore genes	Anders *et al.*, 1973a,b
Menidia menidia^c (Atlantic silverside)/sperm	x ray 800 R	Percent embryo mortality was 25 in control and 71 in eggs fertilized with sperm exposed to 800 R	Engel *et al.*, 1965
Poecilia reticulata^b (tropical guppy)/adults	^{60}Co or x ray 1000-2000 rad	Color-pattern mutation rates were low in comparison with conventional specific-locus rates in the mouse; however, very little data were obtained (only 2 mutants at 1000 rad and 1 mutant at 2000 rad from 852 experimental animals)	Purdom and Woodhead, 1973
Cichlasoma nigrofasciatum^b (convict cichlid)/juveniles	x ray 500 R + 500 R, 24 h apart (1000 R)	Aggressive behavior was reduced in F_1 male progeny of the irradiated group	Holzberg and Schroeder, 1972

Table 17. (Continued.)

Organism/lifestage	Radiation regime[a]	Comments	References
Poecilia reticulata[b] (tropical guppy)/juveniles	x ray 500 and 1000 R	In hybrid and inbred lines, litter size increased in the progeny of irradiated parents; the frequencies of still births and post-natal mortality were reduced in all the post-irradiation generations of the inbred line whereas they were increased significantly in the F_2 generation after spermatogonial irradiation at 1000 R in the hybrid line; sex ratio was not affected by irradiation; the incidence of mutations was higher in post-irradiation generations	Schroeder, 1969b
Poecilia reticulata[b] (tropical guppy)/juveniles	x ray 1000 R	Exposures of 1000 R to early spermatogonia did not increase the exchange frequency between sex chromosomes, but irradiation of stem cells provided a significantly higher incidence of crossing-over events	Schroeder, 1969c
Poecilia reticulata[b] (tropical guppy)/juveniles and adults	x ray 1000 R (oogonia and spermatogonia) 2 x 500 R-spermatozoa	Synergistic interaction of recessive radiation-induced mutations was studied; effects in post-irradiation F_2 were greatest after exposure of spermatozoa with 2 x 500 R followed by single doses of 1000 R to spermatogonia and oogonia	Schroeder and Holzberg, 1972
Oryzias latipes[b] (medaka)/adults	x ray 2000-16,000 R	Dominant lethal mutations were assessed by observing embryo mortality in F_1 progeny of irradiated adults; irradiation of sperm was more effective in inducing dominant lethal mutations than irradiation of oocytes; younger oocytes were more sensitive than older ones; effects were observed at lowest exposure tested, 2000 R	Egami and Hyodo-Taguchi, 1973
Fundulus heteroclitus[c] (mummichog)/sperm	^{137}Cs 5000 R/min 500-150,000 R	Embryos developing from sperm exposed to 5000 R were stunted and malformed (>50%); embryos exposed to high exposures were probably haploid and developed by parthenogenesis	Lasner and Rugh, 1962

---

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time). For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Organism is exclusively marine.

Radiation-induced recessive lethal mutations have also been documented in fish populations in White Oak Lake. Mosquitofish populations (Gambusia affinis) irradiated for several generations produced embryos with a significantly higher mortality percentage than those from a control population (Trabalka and Allen, 1977). The dose rate administered to these organisms was estimated to be 0.59 rad/d, although, as with other studies at White Oak Lake, it is known that radionuclides are not the only contaminant in the lake.

Induction of Chromosomal Aberrations in Invertebrates

Induction of chromosomal aberrations in aquatic invertebrates was examined in two sets of studies (Table 18). The first investigations were those of Blaylock (1966a,b and 1973), who examined effects of irradiation on polytene chromosomes of chironomid midge larvae. Chironomus tentans is an insect with aquatic larval stages - the stages that were tested. Polytene chromosomes, which do not require banding to visualize inversions and small deletions, are particular to certain dipteran species. A 5-y study of chromosomal polymorphism in chronically irradiated (0.63 rad/d) populations in White Oak Lake demonstrated the presence of 10 inversions and 1 deletion that were never observed in control populations. These aberrations were observed in only one of the annual collections, and it was concluded that these mutations were eliminated by natural selection. In contrast, six stable endemic inversions were present in control and irradiated populations, and the frequency of these endemic inversions was not increased in the irradiated populations. Although no detailed population studies were conducted, it was known that C. tentans was abundant in White Oak Lake after exposure to radiation of over 1000 times background levels for over 100 generations.

The dose rates required to detect effects in the laboratory were higher than those in the field. Blaylock (1973) exposed C. tentans larvae to tritium (0.01 to 500 μCi/mL for 20 d) and found that aberrations were first detected at dose rates of 38 rad/d with a minimum accumulated dose of 760 rad.

Harrison et al. (1985) and Anderson et al. (in preparation) have made the first attempts to examine chromosomal aberration induction in marine invertebrates. Because no cell lines exist for any marine invertebrate, studies were conducted in vivo using tissue prepared from embryo and juvenile stages of the marine polychaete Neanthes arenaceodentata. Harrison et al. (1985) examined the induction of chromosomal aberrations and SCE in embryos of

Table 18. Induction of chromosomal aberrations in invertebrates from acute and chronic exposure to radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Chironomus tentans[b] (midge)/larvae	White Oak Lake 0.65 rad/d larval period	5-yr study of chromosomal polymorphism in chronically irradiated (**0.63 rad/d**) population of C. tentans demonstrated the presence of 10 inversions and 1 deletion that were never observed in control populations; these aberrations were observed only once and hence were probably eliminated by natural selection; in contrast, 6 stable endemic inversions were present in control and irradiated populations; the frequency of these endemic inversions was not increased in the irradiated population	Blaylock, 1966a,b
Chironomus tentans[b] (midge)/larvae	Tritium 0.01-500 µCi/mL 20 d	Aberrations were detected in salivary gland polytene chromosomes at 125 µCi/mL and greater (760-3050 rad total dose or **38-153 rad/d**)	Blaylock, 1973
Neanthes arenaceodentata[c] (polychaete)/larvae	x ray 70 rad/min 8-380 rad	Increased frequency of aberration with increase in dose; **≥ 200 rad** required to obtain significant change	Harrison et al., 1985
	^{60}Co 0.005 to 12 rad/h 0.1-300 rad	Increased frequency of sister chromatid exchange; significant increase observed at **60 rad**	
Neanthes arenaceodentata[c] (polychaete)/juveniles	^{137}Cs 500 rad/min 200-2000 rad	Increased frequency of chromosomal aberrations in juvenile worms at **250 rad**, the lowest dose yet tested	Anderson et al., in prep

---

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time); for x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author.

[b] Indicates organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Indicates organism is exclusively marine.

N. arenaceodentata from acute irradiation; they found an increased frequency of chromosomal aberrations with an increase in dose. Doses in excess of 200 rad were required to obtain a significant increase in aberration frequency. An increased frequency of SCE with dose was observed, but it dropped off at doses >100 rad; a significant increase was seen at 60 rad. Results of research in progress with juvenile N. arenaceodentata show an aberration frequency of approximately 0.3 per cell after 200-rad doses. In this study, effect levels for the induction of mortality, decreased fecundity, and chromosomal aberrations were compared. It was determined that significant chromosomal aberration induction occurs at doses 2 to 3 orders of magnitude below those causing death and at approximately the same doses as those that cause reduction in fecundity.

Induction of Mutations in Invertebrates

To our knowledge, very little work has been done to examine the induction in aquatic invertebrates of mutations by radiation. One investigator examined induction of sterility in F_1 progeny of irradiated brine shrimp Artemia salina (Table 19). No effects were observed in brine shrimp receiving 1000 R, the lowest exposure tested; 3500 R induced 44% sterility in F_1 males.

In other studies on A. salina (Metalli and Ballardin, 1962, 1972; Ballardin and Metalli, 1965), diploid and tetraploid strains were exposed to 1000 R from an x-ray source, and hatching success was examined as an index of dominant lethality. For the diploid strain, a reduction in hatching success was seen in the second generation as well as the first, presumably due to the expression of recessive lethal mutations. For the tetraploid strain, no such reduction in hatching success occurred in the second generation. An estimated mutation rate of 3.9×10^{-4} gamete^{-1} rad^{-1} was obtained for the diploid strain in the first generation.

Conclusions on Genetics Data

Genotoxic effects of radiation exposure in fish and aquatic invertebrates have just begun to be examined. Yet, these same effects in mammals are a prime concern of mammalian radiobiologists. No genetic studies have addressed chronic irradiation effects on fish, and only one chronic effects study has been conducted on an aquatic invertebrate (Blaylock 1966a,b). Moreover, the

Table 19. Induction of mutations in invertebrates from acute exposure to radiation.

Organism/ lifestage	Radiation regime[a]	Comments	References
Artemia salina[b] (brine shrimp)/ adults	^{137}Cs 507 R/min 1000-10,000 R	44% sterility in F_1 sons of males at 3500 R was observed; 5000 R produced complete sterility whereas 1000 R produced no significant effect; significant effects on survival to adulthood also occurred at 3500 R and above	Squire, 1973

[a] Radiation regime is presented as source, dose rate, and total dose. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is exclusively marine.

invertebrate examined was the freshwater larva of an insect Chironomus tentans. The lowest effect levels observed to date are (1) the observation by Suyama and Etoh (1983) of a significant increase in dicentric yield in cultured lymphocytes of the fish Umbra limi after 50-R exposures and (2) the induction of major eye malformations in trout embryos after 25-R exposures to sperm (McGregor and Newcombe, 1972a). It is important to note that data obtained to date on genotoxic effects indicate that the sensitivity of some aquatic organisms with respect to cell killing and DNA damage is not less than that observed in mammals. No investigators have examined the role of modifying environmental factors on genotoxic effects.

Further analysis of the genetic effects of ionizing radiation on aquatic organisms is needed. However, careful attention must be paid to selecting endpoints and species to be examined. Moreover, greater attention to experimental design and conduct is necessary to improve the standards of some of the cytogenetic work.

Metalli (1979) emphasized the need for relating studies of genetic effects to endpoints such as reproduction to aid in extrapolating and generalizing genetic data. It was also proposed that the analysis of dominant lethal mutations and metaphase analysis of chromosomal aberrations were the endpoints of choice for future study. He had some reservations about these endpoints, however, and stated that under chronic exposure regimes, non-genetic factors such as meiotic delay may affect the results obtained in dominant lethal studies. He also questioned the expense of chromosomal aberration studies, which were the most highly recommended type of study. In response, we suggest that a portion of future cytogenetic research should use the induction of micronucleis as an important endpoint, because it is less time consuming to quantify than chromosomal aberrations (see Fig. 2 for comparing types of effects detected).

Finally, it must be emphasized that further work in this area will require a substantial research commitment. Even when optimal systems are selected, chromosomal aberration and dominant lethal studies are labor intensive. However, the importance of such work is difficult to contest.

COMPARISON OF RADIATION EFFECTS ON AQUATIC ORGANISMS TO THOSE ON TERRESTRIAL ORGANISMS

Although there are not sufficient data on aquatic organisms to determine the threshold dose rates below which effects would not be observed, we can obtain an indication of what these dose rates might be by comparing the results obtained from exposure of terrestrial animals to those obtained on aquatic organisms. In Fig. 4, low-level effects on mammals are contrasted to those on fishes and aquatic invertebrates, and in Table 20, the critical data on low-level effects on aquatic animals and examples of selected low-level effects on various species of terrestrial animals are provided.

MORTALITY AND PATHOPHYSIOLOGY

To our knowledge, no mortality due to acute irradiation below 10 rad (0.1 Gy) occurs in any animal. Excluding tumors, induction of acute pathophysiologic lesions also occurs at higher doses. The significance of such effects in the deep ocean is equivocal. Because of the long latency period for cancer, long-lived species may be the only organisms at risk.

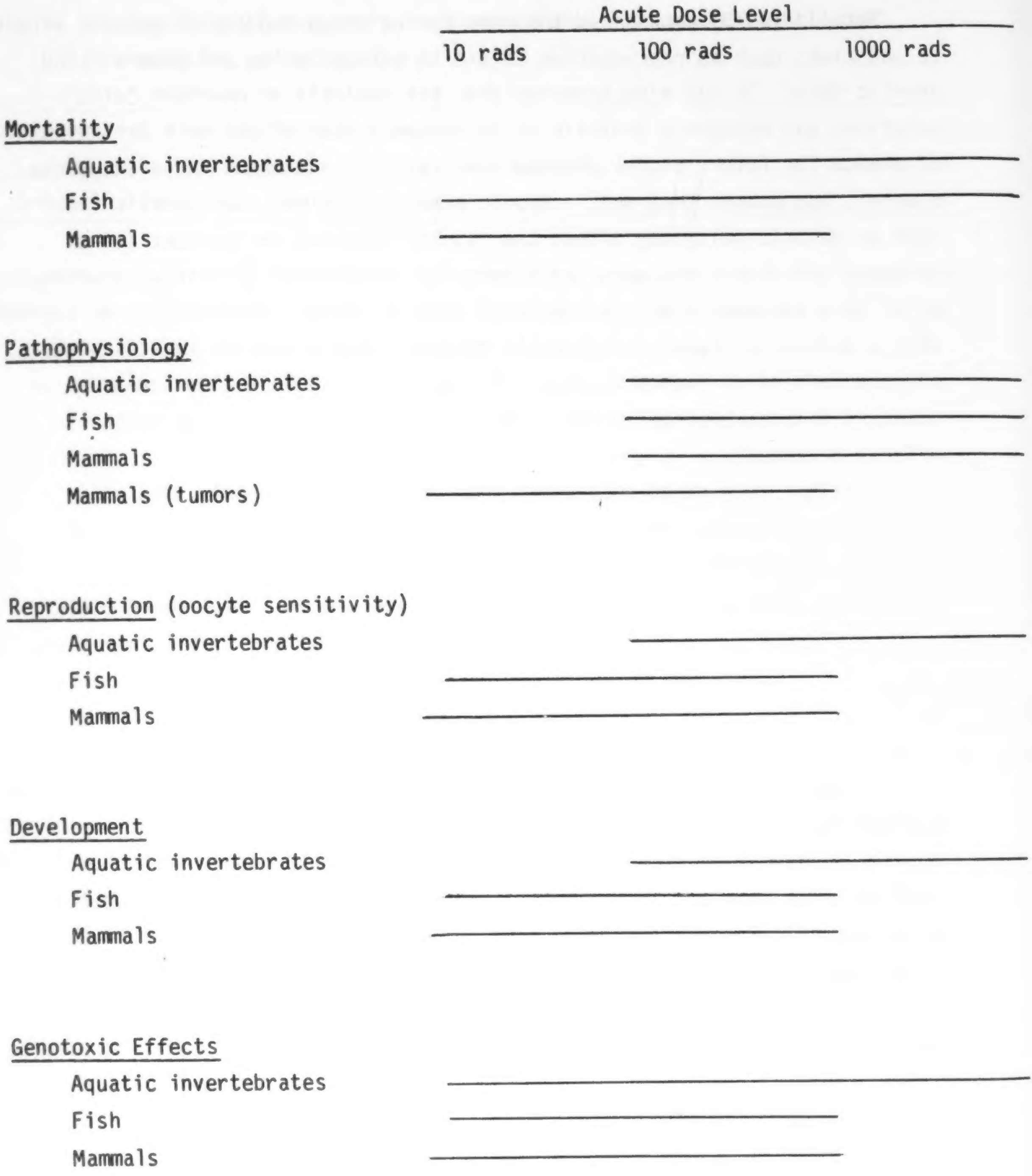


Figure 4. Generalized effect levels for selected biological endpoints.

REPRODUCTION

Gamete death has been demonstrated at low doses in mammals (Fig. 4 and Table 20). It is also known that these response levels are highly species specific. Despite the large amount of controversial data from research on effects of radiation on developing fish embryos, little has been done to define the shape of dose-response curves for gamete killing in fishes and invertebrates. The research that has been done (Table 20) indicates that effects may occur at levels on the order of 1 rad/d (1 x 10^{-2} Gy) or less.

DEVELOPMENT

The production of developmental abnormalities in fish embryos from low doses of radiation has been the topic of considerable debate. Recently, this area of research was reviewed critically by Woodhead (1984). He concluded that effects observed on fish embryos at the lower dose levels from radionuclides in the water are not verifiable. However, embryo death has been induced in fishes with exposures as low as 16 R, and increased developmental abnormalities have been found (with the use of sealed sources) in fish at 0.5 R/d. In contrast to the extensive research on fish, few data are available on the effects of radiation on the development of invertebrates. The data on mammals demonstrate that effects of low levels (3 rad/d) of radiation occur _in utero_ (Table 20) and that they are keyed to critical periods in development.

GENETICS

The significance of effects on genetic material from exposure to low levels of radiation has been debated widely by geneticists. Effects of low-level radiation include increased gene mutation rates, chromosome breakage and rearrangement, and aneuploidy. Production of chromosomal aberrations, which may also result in cell death, is a well-documented effect of radiation and is the result of breakage or combined breakage and exchange of chromosomal material. When stable aberrations such as translocations occur in germ cells, they may be transferred to subsequent generations. A significant increase in chromosomal aberrations in mammalian lymphocytes in culture has been demonstrated in people living in areas with natural high background irradiation (0.11 to 0.34 R/y gamma-ray plus 0.001 to 1.6 R/y alpha ray

Table 20. Summary of biological effects observed at low dose rates and low total doses in aquatic organisms as compared to selected data for terrestrial organisms.

Dose rate or total dose (Source)	Observed or projected effect	Reference
	Aquatic organisms	
0.5 R/d (^{60}Co)	Increased incidence of opercular defects in salmon after 80 d of continuous irradiation beginning immediately after fertilization; a significantly greater average weight in the irradiated group was also observed	Donaldson and Bonham, 1964
0.59 R/d (mixed source, White Oak Lake sediment)	Increased embryo mortality in the mosquitofish *Gambusia affinis* relative to those from an unirradiated field control site; no concomitant decrease in fecundity was observed	Trabalka and Allen, 1977
0.65 R/d (mixed source, White Oak Lake sediment)	Frequency of egg-capsule production was reduced in an irradiated population of the aquatic snail *Physa heterostropha*; however an increased number of eggs per capsule also occurred	Cooley, 1973
4.08 rad/d (^{137}Cs)	Reduced total fecundity at lowest dose tested over the entire life cycle of the guppy *Poecilia reticulata*	Woodhead, 1977
6.8 rad/d (^{137}Cs)	Increased percentage of unfertilized eggs from ricefish *Oryzias latipes* adults irradiated for 120 d; no statistically significant effects were observed at lower doses	Hyodo-Taguchi, 1980
>10 R/d (^{60}Co)	Gonadal development retarded in Chinook salmon smolts irradiated for 80 d after fertilization; lower exposures showed no such effects; the percent return of migrating salmon was slightly higher in the group irradiated at 0.5 R/d	Bonham and Donaldson, 1972; Donaldson and Bonham, 1970

Table 20. (Continued.)

Dose rate or total dose (Source)	Observed or projected effect	Reference
	Aquatic organisms (continued)	
16 R (x ray)	LD_{50} of silver salmon (*Oncorhynchus kisutch*) embryos irradiated in the one-cell stage and observed 150 d after irradiation	Bonham and Welander, 1963
25 rad (^{60}Co)	Increased frequency of major eye malformations in trout embryos from irradiation of trout sperm	McGregor and Newcombe, 1972a
25 and 50 rad (^{60}Co)	Trout sperm at 25 and 50 rad produced decreased embryo mortality; increased embryo mortality from 200-400 rad to sperm	McGregor and Newcombe, 1972b
	Terrestrial organisms[a,b]	
0.11-0.31 R (γ rays) plus 0.001-1.6 R (α rays)	Increased frequency of chromosomal aberrations in human lymphocytes	Pohl-Ruhling and Fischer, 1979
0.043-0.43 R/d (γ rays)	Increased percent sterility in male dogs exposed to 0.043 or 0.086 R/d compared to control; whereas exposure to 0.43 R/d made male dogs sterile in one year	Luckey, 1980
0.3 rad/d (HTO)	Decreased brain weight in F_2 generation of rats irradiated from conception to term	NRC, 1980
1 R/d (γ rays)	Decrease in percent survival in first 6 months of life in desert rodent *Perognathus formosus*; thereafter, percent survival was increased relative to controls	Whicker and Schultz, 1982
1 R/d (^{137}Cs)	Significant increase in mutation rates in mouse spermatogonia versus controls	UNSCEAR, 1977

Table 20. (Continued.)

Dose rate or total dose (Source)	Observed or projected effect	Reference
	Terrestrial Organisms (Continued)	
1.1-1.37 rad/d (γ rays)	Sterility induced in leopard lizard and whiptailed lizard from lifetime irradiation	Whicker and Schultz, 1982
2 rad/d (γ rays)	No significant differences in sex ratios or egg distributions observed in populations of the the lizard *Uta stansburiana*	Whicker and Schultz, 1982
2.5 R/d (^{60}Co)	Prenatal and postnatal mortality observed in rats irradiated from conception to term	NRC, 1980
3 rad/d (HTO)	Reduced brain, testis, and ovary weights in rats irradiated from conception to term	NRC, 1980
3.3 rad/d (HTO)	No effect on lifespan of rats irradiated from conception to term	NRC, 1980
5 rad (HTO) and 8 R (x ray)	LD_{50} of mouse oocytes	NRC, 1980
8 rad (HTO)	LD_{50} of monkey oocytes	NRC, 1980
8.4 R/d (^{137}Cs)	Complete sterilization of female mice irradiated 20-40 d postconception	NRC, 1980

---

[a] Examples selected to represent the range of organisms, effects, and effect levels examined, as well as to illustrate the presence of positive and negative effects at low dose rates. Other examples for terrestrial organisms are given in Luckey, (1980); NRC (1980); UNSCEAR (1977); UNSCEAR (1982); and Whicker and Schultz (1982).

[b] Many examples selected for the section on terrestrial organisms are cited from secondary references.

exposure; Pohl-Ruhling and Fischer, 1979) and an increased dicentric yield in cultured fish lymphocytes was demonstrated at 50 R, the lowest exposure tested. Genotoxic effects of radiation may result in decreases in reproductive success of an organism due to gamete killing and dominant lethal mutations.

LOW-LEVEL RADIATION EFFECTS AND HUMAN HEALTH

The major somatic and heritable genetic effects of low-level radiation have been identified for mammalian populations (NRC, 1980; UNSCEAR, 1977; UNSCEAR, 1982). The principal effect of low-level radiation in somatic tissue is probably cancer induction in a variety of organs and tissues. Also, gamete death and developmental abnormalities to the embryo and fetus have also been observed at low doses, below 10 rad.

Concern over effects on germ cells of low-level radiation centers on the induction of increased frequency of heritable genetic disease. This may occur from mutation, chromosome breakage, chromosome rearrangement, and faulty segregation of chromosomes at metaphase which may result in aneuploidy. The significance of increased frequency of neutral mutations is also the subject of considerable debate.

CONCLUSIONS ON EFFECT LEVELS

Examination of the data on biological effects in aquatic invertebrates, fishes, and mammals at dose rates of approximately 0.5 to 10 rad/d (5×10^{-3} to 1×10^{-1} Gy/d) indicates that those dose rates may define a range in which a variety of low-level effects on reproduction, development and genetic integrity are detectable in sensitive tissues and organisms. It is possible that this range may encompass threshold dose rates for important non-stochastic effects. Furthermore, for aquatic organisms, it is apparent that little information exists that is relevant to the role of factors modifying response levels. Factors such as temperature, species specificity, and cell kinetics may alter observable-effect levels by more than an order of magnitude and thus affect the certainty of predicted effects. Finally, it should be emphasized that the dose range at which data on effects are available is higher than doses measured at currently existing radioactive waste-disposal sites, but the low end of this dose range is only twice the highest dose rates recorded in the ocean due to radioactive waste disposal. Our section on population effects includes a discussion of the importance of decreasing error in effect estimates and the utility of single-species toxicity tests in assessing environmental hazards.

EFFECTS ON POPULATIONS AND COMMUNITIES FROM CHRONIC EXPOSURE TO IONIZING RADIATION

It is stated repeatedly in the aquatic radiobiology literature (Templeton, 1976a; Whicker and Schultz, 1982; Woodhead, 1984) that effects on individuals are not as important for aquatic species as they are for humans. This is certainly true, but it must be remembered that it is general practice in aquatic toxicology that no-effect levels are first determined at the organismal level. That is, discharge criteria and pollution standards are set from effect levels determined in single-species toxicity tests. These effect levels are generally results from standard assays measuring mortality and reproductive effects. Compensation arguments, such as those used in the aquatic radiobiology literature (e.g., that the death of a percentage of a brood is insignificant if the remaining percentage can support a population), are important points of discussion, but, to our knowledge, they are not generally used to set environmental criteria and standards.

VALUE OF SINGLE-SPECIES TOXICITY TESTS IN THE EVALUATION OF EFFECTS ON POPULATIONS AND COMMUNITIES

The adequacy of single-species toxicity tests in estimating environmental hazard is one of the most important considerations in ecotoxicology. On this subject, Cairns (1983) recently concluded that there has not yet been sufficient study to determine the degree of reliability of these tests. He believes there is a great need for more testing at the community level, but also states that single-species toxicity tests are presently the major and only reliable means of estimating environmental hazard.

Cairns (1983) presented a diagram (Fig. 5) showing how the error about an environmental standard or criteria is decreased as more steps are taken to test for no-effect levels. Multiple levels of testing are not required when expected environmental concentrations are much lower than no-effect levels, unless, for some reason, an extremely low level of error is necessary. This

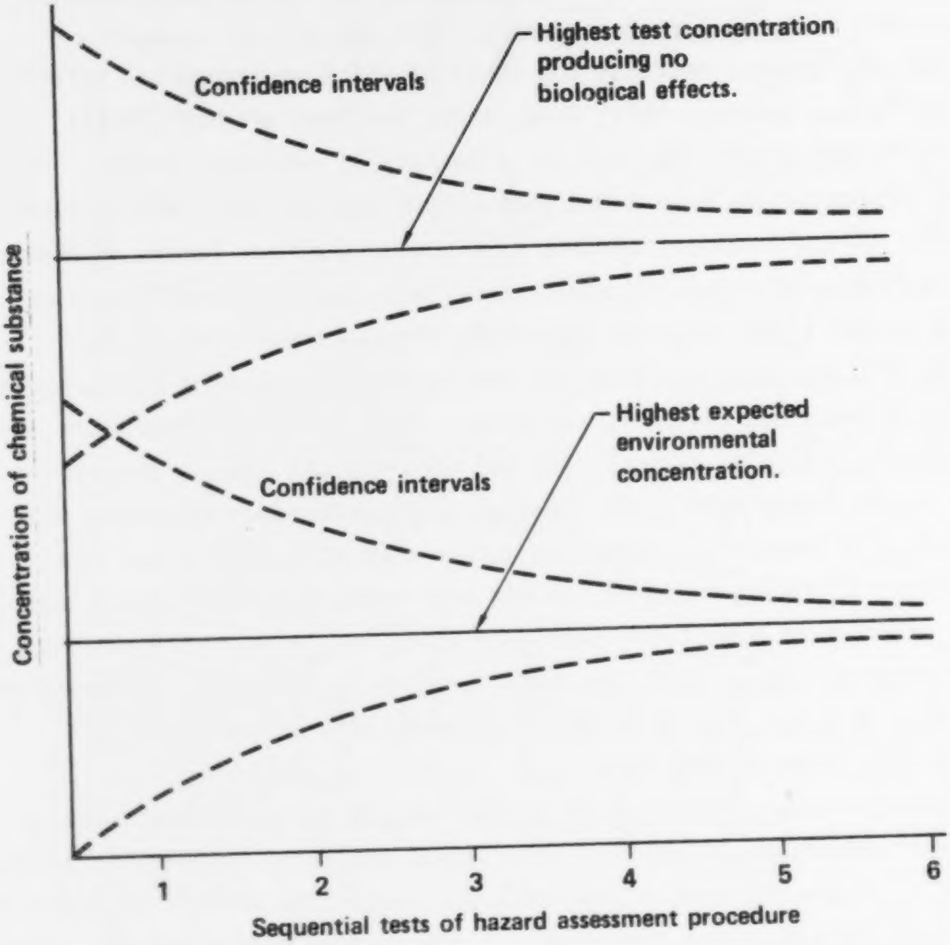


Figure 5. The relationship between error of effect-level estimates and stages of investigation (Cairns, 1983). The dashed lines represent confidence intervals around the line representing the highest expected concentration.

is probably the situation that exists with regard to dumping low-level wastes in the ocean. The recent NACOA report (1984) emphasized the need for societal consensus on nuclear waste management and its effect on decision making regarding dumping radioactive wastes into the ocean.

Many investigators performing ecotoxicological research have argued that single-species toxicity tests are inadequate for estimating environmental hazard and that ecological factors such as food density and community interactions may greatly decrease the level at which environmental effects can be detected (e.g., Cushing, 1979; Gray, 1979; Kooijman and Metz, 1984). Perkins (1979) has argued the need for a variety of sublethal tests.

Other investigators have found that single-species tests are predictive of community effects. Recent studies have compared effect levels of chemical contaminants using organismal-, population-, and community-level endpoints. Hansen and Garton (1982) exposed laboratory stream communities to the insecticide diflubenzuron for 5 months and assessed changes at the community level using biomass and diversity estimates. They then conducted acute and chronic toxicity tests on several fish and invertebrate species present in the streams. It was found that single-species tests adequately predicted the diflubenzuron concentrations affecting stream communities, but that the concentrations eliciting responses in the most sensitive test species were more than an order of magnitude lower than those resulting in the observed community effects. Because of the short duration of the tests on communities, however, they were not able to assess the sensitivity of important population-level reproductive effects.

A recent EPA study (Mount *et al*., 1984) showed that toxicity testing using ambient waters was predictive of instream community response to pollution in the Ottawa River. In this study, fish and benthic invertebrate samples were taken at nine stations along the river. A sewage-treatment plant was located between stations 2 and 3, a refinery between stations 3 and 4, and a chemical plant between 4 and 5. In parallel, water samples were taken at these stations, and the water was used to run reproductive bioassays on *Ceriodaphnia sp*. A similar pattern (Fig. 6) was found between number of benthic species represented at a station and the number of young per female *Ceriodaphnia sp*. obtained in bioassays. This is a particularly interesting result because the most classical combined organismal and population studies in the aquatic radiobiology literature were conducted on *Daphnia pulex*; these studies are described in the next section.

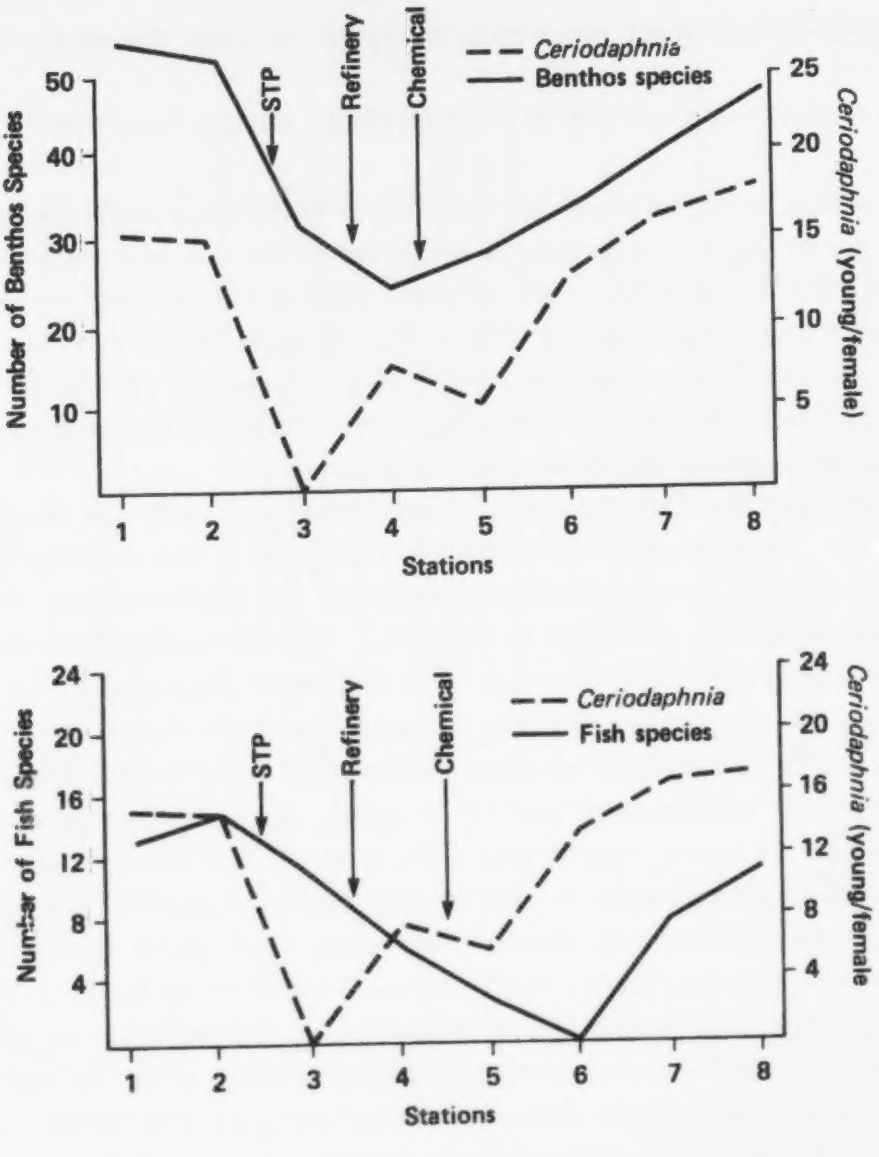


Figure 6. Ambient toxicity correlation between Ceriodaphnia young per female and ecological survey data (Mount et al., 1984).

In summary, there is as yet no consensus on the use of community-level testing in aquatic toxicology, and these tests are generally considered as second-tier tests, to be used after no-effect levels are determined at the organismal level. Therefore, it is concluded that determination of no-effect levels using relevant organismal-level endpoints, especially reproduction, should remain the highest research priority for at least the next few years.

EFFECTS OF IONIZING RADIATION ON POPULATIONS OF AQUATIC ORGANISMS

A variety of perspectives on the effects of ionizing radiation on populations of aquatic organisms is presented in reviews by Templeton (1976a), Blaylock and Trabalka (1978), and Woodhead (1984). In a recent review, Whicker and Schultz (1982) examined the effects of radiation on terrestrial and aquatic populations. In this review, we will briefly discuss the results of population-level studies on aquatic organisms (Table 21) and compare effect levels to those observed for terrestrial organisms.

The classic studies of Marshall (1962, 1966, and 1967) are the first and only studies in the aquatic radiobiology literature to use quantitative population biology to study radiation effects. The basic findings of these extensive studies are presented in Table 21. Marshall (1962) determined that the dose rate at which the intrinsic rate of natural increase, r, was reduced in chronically irradiated _Daphnia_ _pulex_ populations was 70 R/h. Birth rate fell sharply at 50 R/h, whereas death rate was barely affected at these dose rates. Marshall (1966) found that for _D. pulex_ populations chronically irradiated for 55 weeks, populations exposed to 436 R/d or more became extinct. Marshall (1962) established that the maximum tolerable exposure rate was 1330 R/d. The difference in response was due to food-supply limitation in the second study. In a subsequent study, Marshall (1967) examined the effects of exploitation on food-limited, irradiated _D. pulex_ populations. He found that, up to a certain level, exploitation had an ameliorating effect on radiation stress. These studies would seem to indicate that very high chronic exposure rates might be tolerated by invertebrate populations. However, evidence for reproductive and genetic effects below these levels has been given by several authors.

Studies conducted at White Oak Lake, which have been discussed in other sections, demonstrated reproductive effects in snail and fish populations at dose rates below 1 rad/d (Cooley, 1973; Trabalka and Allen, 1977) and genetic

Table 21. Induction of effects on populations of aquatic organisms from chronic exposure to radiation. Entries are ordered according to the lowest dose at which effects were observed. This dose is not necessarily the lowest dose at which effects could have been observed.

Organism/lifestage	Radiation regime[a]	Comments	References
Oncorhynchus tschawytscha[b] (chinook salmon)/embryos	^{60}Co 0.5-1.3 R/d 40-104 R	At **0.5 R/d** slightly higher rates of return in migrating salmon were observed	Bonham and Donaldson 1966; Donaldson and Bonham, 1970
Gambusia affinis[b] (mosquitofish)/all stages	White Oak Lake 0.59 rad/d lifetime	No decrease in fecundity observed at **0.59 rad/d**; a significant increase in frequency of dead embryos in the irradiated population versus control was demonstrated; this indicated the presence of radiation-induced recessive lethal mutations maintained in the population since initial studies in 1966	Trabalka and Allen, 1977
Chironomus tentans[b] (midge)/larvae	White Oak Lake 0.65 rad/d larval period	5-y study of chromosomal polymorphism in chronically irradiated (**0.65 rad/d**) population of *C. tentans* demonstrated the presence of ten inversions and one deletion that were never observed in control populations; these aberrations were observed only once and hence were probably eliminated by natural selection; in contrast, 6 stable endemic inversions were present in control and irradiated populations; the frequency of these endemic inversions was not increased in the irradiated population	Blaylock, 1966a,b
Physa heterostropha[b] (aquatic snail)/adults	White Oak Lake 0.65 rad/d lifetime	Frequency of egg-capsule production was reduced in the irradiated (**0.65 rad/d**) population, but an increase in number of eggs per capsule occurred, resulting in similar rates of egg production compared to controls	Cooley, 1973
Daphnia pulex[b] (daphnia)/complete life cycle	^{60}Co 0-516 R/d; populations irradiated 18.5 h/d for 55 weeks	Populations exposed to **436 R/d** or more became extinct	Marshall, 1966

Table 21. (Continued.)

Organism/lifestage	Radiation regime[a]	Comments	References
Daphnia pulex[b] (daphnia)/complete life cycle	^{60}Co 22.8-75.9 R/h 19 h/d 20-35 d	Exposure rate at which intrinsic natural rate of increase was reduced to zero was **70 R/h**; birth rate fell sharply at **50 R/h**	Marshall, 1962
Daphnia pulex[b] (daphnia)/complete life cycle	x ray 3.7-5.1, 108-162, 270-378, and 468-684 R/d 80-100 weeks	Effects of exploitation on food-limited D. pulex were studied; exploitation decreased radiation effects observed in food-limited populations	Marshall, 1967
Artemia salina[c] (brine shrimp)/complete life cycle	x ray 1000, 2500, 3000 R/ few min (irradiated 3 times per y for 2 y)	Culture survival only at **1000 R**; no life-span change at 1000 R, but decrease in number of zygotes voided and number of mature adults per pair	Grosch, 1962

---

[a] Radiation regime is presented as source, dose rate, and total dose (or exposure time). For x-ray data, a dose rate is not given; factors important in determining x-ray dose rate are voltage, target material, filtration, tube current, and target-to-object distance. Units are those used by the author; R is the abbreviation for roentgen.

[b] Organism is freshwater, anadromous, or estuarine as opposed to exclusively marine.

[c] Organism is exclusively marine.

effects in Chironomus tentans populations at similar dose rates (Blaylock, 1966a,b). Effects at these low levels have not been determined in the laboratory. However, reproductive effects between 1 and 10 rad/d have been demonstrated in the laboratory (Woodhead, 1977; Hyodo-Taguchi, 1980).

Templeton (1976a) has stated that highly fecund fish species with rapid development rates would be at less risk to radiation exposure than low-fecundity fish species with slow development rates. This hypothesis has not been tested on fish, but Turner et al. (1973) tested this hypothesis in populations of lizards chronically irradiated in the field. Some species of lizards became sterile after long-term irradiation at 1.1 to 1.3 R/d, whereas other lizard species were unaffected by the same radiation exposure. These differences in responses of the populations were attributed to differences in population dynamics rather than differences in gamete sensitivity. It was found that slow-developing, low-fecundity species were most affected by chronic irradiation because higher doses could be accumulated. Sterility has also been induced in dogs with chronic exposures of 0.43 R/d (Luckey, 1980).

Much evidence exists for induction of hormesis from low levels of ionizing radiation (Luckey, 1980). Intriguing results (Donaldson and Bonham, 1970), indicating an increased return rate of salmon that had been irradiated as embryos at 0.5 R/d versus unirradiated controls, are frequently given as evidence of hormesis. However, in the same series of studies, Donaldson and Bonham (1964) demonstrated an increase in opercular defects in embryos at the same dose rate.

PROVISIONAL DOSE ASSESSMENTS FOR DEEP-SEA ANIMALS

In risk-assessment procedures, knowledge of potential biological effects and effect levels for a contaminant are combined with information about the fate of the contaminant in the environment and within organisms. This report focuses on the biological effects of low-level radiation on aquatic organisms. However, currently used provisional dose assessments for deep-sea animals are compared in this section to the biological effects levels we have described.

Woodhead and Pentreath (1983) made an assessment of the doses that may be received from natural background radiation by deep-sea organisms (Table 22) and compared these doses to those received by coastal organisms. Their calculations indicate that the natural radiation background of deep-sea organisms is likely to be as high and at least as variable as that of coastal

Table 22. Estimates of the radiation dose rates (nSv/h) to benthic deep-sea organisms from natural background and the peak dose rates predicted from dumping low-level, radioactive wastes (OECD/NEA, 1985).[a]

	Fish	Large crustaceans	Molluscs	Small crustaceans
Background	8.5×10^1 to 1.4×10^3	9.9×10^2 to 4.6×10^3	9.3×10^2 to 3.4×10^3	3.3×10^2 to 4.2×10^3
Past dumping	7.3×10^1	3.8×10^2	8.0×10^3	1.3×10^3
Past + future dumping	1.0×10^2	5.0×10^2	1.0×10^4	1.7×10^3
Past + future dumping x 10	6.2×10^2	2.9×10^3	6.5×10^4	9.7×10^3

[a] Radiation dose based on the assumption that the radionuclides are deposited in a rectangular box 40 km x 120 km x 75 m deep and include the underlying sediments.

and shallow-water organisms, and they concluded that deep-sea organisms may have evolved, therefore, in a radiation regime similar to that of coastal organisms. However, extrapolation of results of radiation effects on coastal organisms to those on deep-ocean organisms should be made with caution for the following reasons:

1. Models used to estimate the radiation doses to deep-ocean organisms were simplistic because insufficient data were available on radionuclide (particularly alpha emitters) distribution in the tissues (particularly gonadal tissue) of the organisms.

2. Information on the dose-effect relationships for different classes of deep-ocean organisms is not available. Therefore, it is not known how the effect of dose would be modified by such environmental factors as low temperature, low oxygen tension, high pressure, etc.
3. Evolution in a similar radiation regime may not result in similar radiosensitivity. For example, the mouse and the rat have evolved in similar radiation regimes, but the sensitivity of the rat and mouse oocytes to radiation differ significantly.

As more information becomes available on the internal distribution within deep-sea biota and the external distribution in the environment of radionuclides of concern, model predictions can be refined, and the conservatism required to ensure the protection of the environment can be reduced.

A considerable amount of attention has been given to the prediction of the dose to organisms indigenous to the Northeast Atlantic Disposal Site, which has been receiving low-level radioactive waste since 1949. The feasibility of the continued use of this site has been under evaluation (OECD/NEA, 1985). Radiation doses to bathypelagic and benthic deep-sea organisms have been calculated assuming that the radionuclides are deposited in a rectangular box 40 km x 120 km x 75 m deep, a box 250 km x 250 km x 500 m, and a box 2500 km x 3500 km x 1000 m deep. The boxes include the underlying sediment. Results of the calculations for the smallest box and for benthic organisms are given in Table 22. These results are the highest of the three and their use would provide conservative estimates of potential detrimental impact.

The highest dose rates predicted are those received by the benthic molluscs at the site; these are greater than the maximum estimated for natural background. As a result of using the most conservative model, the results of calculations indicate that benthic molluscs would be exposed to 6.5×10^4 nSv/h or 0.156 rem/d. According to data we have tabulated, this dose rate is approximately an order of magnitude below the level at which the most subtle reproductive and genotoxic effects are noted.

ADDITIONAL METHODOLOGIES FOR THE STUDY OF RADIOBIOLOGICAL EFFECTS

Because the effects of radiation exposure in mammals have been of widespread concern, many methodologies for studying radiobiological effects on genetic material have been developed. Some of these approaches have never been applied to the study of effects on aquatic organisms, and three, in particular, may provide very useful information.

Analyzing the quantity of DNA single-strand breaks caused by radiation exposure is possible because damaged DNA unwinds more rapidly than intact DNA when cell lysates are exposed to alkaline conditions. DNA unwinding in treated-cell lysates is monitored on a fluorometer. By using this and similar techniques on appropriate tissues from aquatic organisms, investigators may be able to detect effects from radiation exposures as low as 5 to 10 rad or 0.05 to 0.1 Gy (Birnboim and Jevcak, 1981). Furthermore, the relative rates of strand breakage and repair could be studied in different aquatic organisms, provided appropriate tissues are identified. The drawbacks of research in this area are (1) that two to three years of development would be required to adapt the technique and to identify the best tissues for study and (2) that a large proportion of DNA single-strand breaks are repaired (hence both repair and recovery would have to be studied).

Unscheduled DNA synthesis (UDS), studied as an indicator of DNA repair activity, is generally measured by analyzing ^{3}H-thymidine uptake in exposed cells or tissues. Uptake is usually quantified by autoradiography, although other methods exist. Recently, Tuschl et al. (1983) reported increased rates of UDS in lymphocytes of persons occupationally exposed to greater than 0.014 rad/month over background levels. Development of this technique could aid in the detection of genetic effects from low-level radiation exposure as well as provide means for comparing relative rates of DNA repair among aquatic organisms and between aquatic organisms and mammals. A minimum of two to three years of development would be required before these kinds of comparisons could be addressed.

The third technique, premature chromosome condensation, involves the fusion of interphase cells from a test organism with a second type of cell that is rapidly dividing. The effect is that interphase chromosomes may be induced to condense prematurely such that chromosomal aberrations can be visualized (Pantelias and Maillie, 1983). This technique has distinct relevance to invertebrate radiobiology because it enables the investigator to directly analyze chromosomal damage without complications in interpretation arising from interphase death of cells, mitotic delay, or cell repopulation. The first step in the development of such an approach would be the selection of appropriate tissues for fusion. This is a difficult first step because, to our knowledge, fusion of marine invertebrate cells has only been conducted on sea urchin embryos.

Other important assays of DNA damage exist that have not been mentioned here. Developing any of these assays for application to aquatic organisms will require a commitment to basic research before meaningful answers are provided. However, such fascinating and important basic issues (which also have practical predictive value) as the relative abilities of different members of the animal kingdom to repair DNA await analysis.

MONITORING APPROACHES

The purpose of this review has been to examine the current state of knowledge relevant to determining the potential biological effects of low levels of ionizing radiation on aquatic organisms. The information provided can be used by others to evaluate the potential hazard of dumping low-level, radioactive waste into the ocean. An integral part of aquatic toxicological hazard assessment is the determination of NOELs for a contaminant under study. Exposure values are then incorporated into models to predict the impact on a specific area of a given contaminant concentration. Next, cost-benefit analyses are made to weigh the social and economic values of the activity against the environmental risks. If a decision to resume dumping of low-level radioactive wastes in the ocean is made on the basis of knowledge of the effect levels, then a monitoring strategy would serve to check the accuracy of predicted effects. Thus, development of monitoring methodologies is a secondary goal, whereas the determination of no-effect or minimal-effect levels is a primary goal in the decision-making process.

There are two general approaches to monitoring. The first is to directly examine communities in impacted and control areas to determine whether significant effects have occurred in the impacted area. Unfortunately, the low biomass, high diversity, high patchiness, and extreme difficulty of doing routine sampling in the deep sea make this option unfeasible. The second approach to monitoring is to determine the potential mechanisms for impact on individual organisms and try to relate the magnitude of an observed response to potential detrimental effects. To develop such an environmental dosimeter for marine organisms, an endpoint of significance must be studied in a variety of organisms under a variety of conditions to determine its potential sensitivity and reproducibility. Furthermore, effect levels must be related to other biological endpoints to determine the risk associated with any given exposure.

The prime method for biological dosimetry due to radiation exposure in human populations is metaphase analysis of chromosomal aberrations in lymphocytes. Use of this analysis helps reconstruct the exposure level of the person being examined. When this is linked with other knowledge about the biological effects of such an exposure, biological risk estimates for this person can be made.

To translate this method of environmental dosimetry to marine organisms, considerable research on the basic cell biology of the organisms and tissue in question is required. An understanding of the cell kinetics of a given system is essential because a certain percentage of aberrations are lost at each cell division and because radiation induces mitotic delay that affects the time at which damage is seen. Other complicating factors such as interphase death of damaged cells may also occur.

A second, very recent approach to biological dosimetry is the use of the production from radiation of electron paramagnetic resonance (EPR) signals in calcified tissue (McCreery et al., 1984). The nature of the radiation-induced EPR signal in calcified tissues makes it amenable for use in biologic dosimetry. This signal is associated with lattice defects or electron traps within calcified material and has been examined in human bone and teeth that have been irradiated in vitro. The signal is extremely stable and, at normal temperatures, the time constant is approximately a million years. The response to radiation appears to be dose related; the sensitivity is down to a total dose of 10 to 20 rads; and data can be obtained with samples as small as 10 to 20 mg. This technique has been proposed for evaluating radiation exposure to military personnel. However, it should be equally applicable for detecting radiation exposure of bone or teeth from deep-sea organisms or calcified exoskeletons from deep-sea invertebrates. Of special interest would be the response in brittle-star exoskeletons because brittle stars live in and on the sediment layer, which would optimize their exposure to radiation.

best copy available

RECOMMENDATIONS FOR FUTURE RESEARCH ON BIOLOGICAL EFFECTS AND BIOMONITORING STRATEGIES

Even though a large body of literature exists on the effects of ionizing radiation on aquatic organisms, only a fraction of it addresses the specific needs of decision makers with regard to ocean dumping of radioactive waste.

The general deficiencies in the data base are (1) few reliable studies at chronic and low dose rates have been conducted, (2) few studies have been done on marine organisms (which means that the comparative sensitivity of these species with regard to selected endpoints is unknown), (3) very few studies have been done on marine invertebrates, (4) information on modifying factors such as temperature, species specificity, and cell kinetics is very scarce, and (5) the long-term effects of low-level radiation on fertility in fish and invertebrates have not been adequately characterized.

To characterize low- or threshold-effect levels in a time-effective and cost-effective manner, the most relevant biological endpoints, species, and modifying factors should be identified. Our recommendations are as follows:

1. In view of evidence for radiation-induced sterility at long-term low doses in both fish and mammals, further studies should be conducted to determine gamete sensitivity and frequency of dominant lethal mutations in a variety of species of marine fishes and invertebrates.

2. Information is needed on the effects of chronic irradiation on marine invertebrates. This information should relate to parallel or previous acute studies so that acute:chronic ratios can be determined.

3. Results from research indicate that analysis of chromosomal aberrations in fish and invertebrates may be a more sensitive endpoint than gamete death. For this reason, further studies should be conducted to characterize chromosomal aberration induction with regard to the sensitivity of various species and the cell kinetics of specific tissues. Use of the micronucleus assay should also be considered.

4. Because temperature can modify biological responses to radiation by more than an order of magnitude (causing fractionated or chronic doses to be more clearly additive) and because nothing has been done on this effect in cold-adapted marine organisms, research should be done to examine the role of low temperature on radiation damage at low-level, chronic doses. Species that normally live in cold environments should be selected, and a variety of endpoints, including oocyte sensitivity, observed at different chronic and acute doses to examine the possibility of a decrease in the dose-rate effect.

If biological monitoring of ocean disposal sites becomes a high priority, a research effort should be considered to develop and characterize a method of biological dosimetry beyond the determination of body burdens. Because studying parameters of community structure or population reproduction are currently unfeasible in the deep ocean, we have sought other options and identified two potential approaches:

First, examination of chromosomal aberrations in selected organisms has potential for environmental dosimetry. However, development of this technique with regard to ultimate sensitivity, reproducibility, and species selection is a long-term project. In some cases, such an effort may be combined with the reproductive studies recommended above to provide important information on the relative sensitivities of these endpoints.

The second option identified is the application of electron paramagnetic resonance (EPR) spectroscopy to evaluate radiation effects in calcified tissues. The nature of radiation-induced EPR signal in calcified tissues makes it amenable for use as a biologic dosimeter. It is proposed that calcified exoskeletons of invertebrates or bones of fishes be used as the test material.

ACKNOWLEDGMENTS

The authors wish to acknowledge M. Varela and R. Dyer at the U.S. Environmental Protection Agency, Office of Radiation Programs for their assistance in the development of this manuscript. Others who provided scientific advice are also gratefully acknowledged, they are: L. Anspaugh, B. Backus, A. V. Carrano, N. Nelson, and T. Straume. In addition, B. Strack and G. Reed provided editorial assistance, and A. Fountain and J. Johansen produced the manuscript.

REFERENCES AND BIBLIOGRAPHY

Abrahamson, S., M.A. Bender, A.D. Conger, and S. Wolff (1973), "Uniformity of Radiation-Induced Mutation Rates Among Different Species," Nature 245, 460.

Achey, P.M., A.D. Woodhead, and R.B. Setlow (1979), "Photoreactivation of Pyrimidine Dimers in DNA from Thyroid Cells of the Teleost, *Poecilia formosa*," Photochem. Photobiol. 29, 305.

Allen, A.L., and Mulkay, L.M. (1960), "X-Ray Effects on Embryos of the Paradise Fish with Notes on the Normal Stages," Growth 24, 131.

Anders, A., F. Anders, and K. Klinke (1973a), "Regulation of Gene Expression in the Gordon-Kosswig Melanoma System: I. The Distribution of the Controlling Genes in the Genome of the Xiphophorin Fish, *Platypoecilus maculatus* and *Platypoecilus variatus*," in Proc. Symp. Genetics and Mutagenesis of Fish, J.H. Schroeder, Ed. (Springer-Verlag, New York, NY), pp. 33-52.

Anders, A., F. Anders, and K. Klinke (1973b), "Regulation of Gene Expression in the Gordon-Kosswig Melanoma System: II. The Arrangement of Chromatophore Determining Loci and Regulating Elements in the Sex Chromosomes of Xiphophorin Fish, *Platypoecilus maculatus* and *Platypoecilus variatus*," in Proc. Symp. Genetics and Mutagenesis of Fish, J.H. Schroeder, Ed. (Springer-Verlag, New York, NY), pp. 53-63.

Anderson, S.L., F.L. Harrison and G. Chan (in prep.), "Chromosomal Aberrations, Reduced Fecundity and Lifespan in Irradiated Polychaete Worms."

Angelovic, J.W., and D.W. Engel (1968), "Interaction of Gamma Irradiation and Salinity on Respiration of Brine Shrimp (*Artemia salina*) Nauplii," Radiat. Res. 35, 102.

Angelovic, J.W., J.C. White, Jr., and E.M. Davis (1969), "Interactions of Ionizing Radiation and Temperature on the Estuarine Fish, *Fundulus heteroclitus*," in *Proc. 2nd Natl. Symp. Radioecology*, CONF-670503, D.J. Nelson and F.C. Evans, Eds. (U.S. Atomic Energy Commission, Washington, DC), pp. 131-141.

Aoki, K., N. Egami, and R. Arai (1966), "Histological Changes in Interrenal Tissue of the Goldfish, *Carassius auratus*, following X-Irradiation," *J. Radiat. Res.* *7*, 83.

Arena, V., Ed. (1971), *Ionizing Radiation and Life* (The C.V. Mosby Company, St. Louis, MO), 543 p.

Ayala, F.J. (1966), "Evolution of Fitness. I. Improvement in the Productivity and Size of Irradiated Populations for *Drosophila serrata* and *Drosophila birchii*," *Genetics* *53*, 883.

Bacq, Z.M., and P. Alexander (1961), *Fundamentals of Radiobiology* (Pergamon Press, Oxford), 562 p.

Ballardin, E., and P. Metalli (1965), "Sulla Relazione Fra Poliploidia e Radiosensibilita in Ovociti de *Artemia salina*, Leach," *Boll. Zool.* *32*, 613.

Baptist, J.P., D.A. Wolfe, and D.R. Colby (1976), "Effects of Chronic Gamma Radiation on the Growth and Survival of Juvenile Clams (*Mercenaria mercenaria*) and scallops (*Argopecten irradians*)", *Health Phys.* *30*, 79.

Bedford, J.S. (1982), "Repair of Sublethal Damage and the Dose-Rate Effect," in *Radiation Biology*, D.J. Pizzarello, Ed. (CRC Press, Inc., Boca Raton, FL), pp. 70-81.

Bierman, V.J., Jr., J.H. Gentile, J.F. Paul, D.C. Miller, and W.A. Brungs (1984), *Research Strategy for Ocean Disposal: Conceptual Framework and Case Study*, Society of Environmental Toxicology and Chemistry, Special Publication No. 1, (in press).

Birnboim, H.C. and J.J. Jevcak (1981), "Fluorometric Method for Rapid Detection of DNA Strand Breaks in Human White Blood Cells Produced by Low Doses of Radiation," *Cancer Res.* *41*, 1889.

Blaylock, B.G. (1966a), "Chromosomal Polymorphism in Irradiated Natural Populations of *Chironomus*," *Genetics* *53*, 131.

Blaylock, B.G. (1966b), "Cytogenetic Study of A Natural Population of *Chironomus* Inhabiting an Area Contaminated by Radioactive Waste," in *Proc. Symp. Disposal of Radioactive Wastes into Seas, Oceans, and Surface Waters* (International Atomic Energy Agency, Vienna), pp. 835-846.

Blaylock, B.G. (1969), "The Fecundity of a *Gambusia affinis affinis* Population Exposed to Chronic Environmental Radiation," *Radiat. Res.* *37*, 108.

Blaylock, B.G. (1973), "Chromosome Aberrations in *Chironomus riparius* Developing in Different Concentrations of Tritiated Water," in *Proc. 3rd Natl. Symp. Radioecology*, *CONF-710501*, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1169-1178.

Blaylock, B.G. and M.L. Frank (1980), "Effects of Chronic Low-Level Irradiation on *Gambusia affinis*," in *Radiation Effects on Aquatic Organisms*, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 81-90.

Blaylock, B.G. and N.A. Griffith (1971), "Effect of Acute Beta and Gamma Radiation on Developing Embryos of Carp (*Cyprinus carpio*)," *Radiat. Res.* *46*, 99.

Blaylock, B.G. and P.G. Koehler (1969), "Terminal Chromosome Rearrangements in *Chironomus riparius*," *Am. Nat.* *103*, 547.

Blaylock, B.G. and T.J. Mitchell (1969), "The Effect of Temperature on the Dose Response of *Gambusia affinis affinis* from two Natural Populations," *Radiat. Res.* *40*, 503.

Blaylock, B.G. and H.H. Shugart, Jr. (1972), "The Effect of Radiation-Induced Mutations on the Fitness of Drosophila Populations," Genetics 72, 469.

Blaylock, B.G., and J.R. Trabalka (1978), "Evaluating the Effects of Ionizing Radiation on Aquatic Organisms," in Advances in Radiation Biology, Vol. 7, J.T. Lett and H. Adler, Eds. (Academic Press, New York, NY), pp. 103-152.

Blaylock, B.G. and J.P. Witherspoon (1975), "Dose Estimation and Prediction of Radiation Effects on Aquatic Biota Resulting From Radioactive Releases From the Nuclear Fuel Cycle," in Proc. Symp. Impacts of Nuclear Releases into the Aquatic Environment (International Atomic Energy Agency, Vienna), pp. 377-393.

Bonham, K., and L.R. Donaldson (1966), "Low-Level Chronic Irradiation of Salmon Eggs and Alevins," in Proc. Symp. Disposal of Radioactive Wastes into Seas, Oceans, and Surface Waters (International Atomic Energy Agency, Vienna), pp. 869-883.

Bonham, K., and L.R. Donaldson (1972), "Sex Ratios and Retardation of Gonadal Development in Chronically Gamma-Irradiated Chinook Salmon Smolts," Trans. Am. Fish. Soc. 101, 428.

Bonham, K., and A.D. Welander (1963), "Increase in Radioresistance of Fish to Lethal Doses with Advancing Embryonic Development," in Proc. 1st Natl. Symp. Radioecology, V. Schultz and A.W. Klement, Eds. (Reinhold Publishing Corp., New York, NY), pp. 353-358.

Brown, F.A., Jr. (1963), "An Orientational Response to Weak Gamma Radiation," Biol. Bull. 125, 206.

Brown, F.A., Jr. and Y.H. Park (1964), "Seasonal Variations in Sign and Strength of Gamma-Taxis in Planarians," Nature 202, 469.

Brown, V.M., and W.L. Templeton (1964), "Resistance of Fish Embryos to Chronic Irradiation," Nature 203, 1257.

Cabela, E., E. Wawra, and H. Altmann (1972), "Repair of Radiation-Induced DNA Strand Scissions in Chlorella During the Cell Cycle," Biophysics 8, 88.

Cairns, J., Jr. (1983), "Are Single Species Toxicity Tests Alone Adequate for Estimating Environmental Hazard?" Hydrobiologia 100, 47.

Calkins, J. (1964), "The Lethal Effects of Radiation on Six Species of Protozoans," Photochem. Photobiol. 3, 143.

Casarett, A.P. (1968), Radiation Biology (Prentice-Hall, Inc., Englewood Cliffs, NJ), 368 p.

Cervini, A. and S. Giavelli (1965), "Radiosensitivity of Different Meiotic Stages of Oocytes in Parthenogenetic Diploid Artemia salina Leach", Mutat. Res. 2, 452.

Chee, P.C., R.J. Vetter, and G.J. Atchison (1979), "Effect of Dose Rate on Survival Time in the Fathead Minnow, Pimephales promelas," Radiat. Res. 77, 98.

Chipman, W.A. (1972), "Ionizing Radiation," in Marine Ecology, Vol. I, Pt. 3, Ch. 11, O. Kinne, Ed. (John Wiley/Interscience, New York, NY), pp. 1579-1657.

Conte, F.P. (1965), "Effects of Ionizing Radiation on Osmoregulation in Fish Oncorhynchus kisutch," Comp. Biochem. Physiol. 15, 293.

Cooley, J.L. (1973), "Effects of Chronic Environmental Radiation on a Natural Population of the Aquatic Snail Physa heterostropha," Radiat. Res. 54, 130.

Cooley, J.L. and F.L. Miller, Jr. (1971), "Effects of Chronic Irradiation on Laboratory Populations of the Aquatic Snail Physa heterostropha," Radiat. Res. 47, 716.

Cosgrove, G.E. and B.G. Blaylock (1973), "Acute and Chronic Irradiation Effects in Mosquitofish at 15 or 25°C," in *Proc. 3rd Natl. Symp. Radioecology*, *CONF-710501*, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 579-584.

Cosgrove, G.E., B.G. Blaylock, G.U. Ulrikson, and P.H. Cohan (1975), "Radiation-Induced Hematopoietic Lesions in Fish," in *The Pathology of Fishes*, W.E. Ribelin and G. Migaki, Eds. (University of Wisconsin Press, Madison, WI), pp. 463-476.

Crenshaw, J.W. (1965), "Radiation-Induced Increases in Fitness in the Flour Beetle *Tribolium confusum*," *Science* *149*, 426.

Croute, F., J.P. Soleilhavoup, S. Vidal, D. Dupouy, and H. Planel (1982), "*Paramecium tetraurelia* Growth Stimulation under Low-Level Chronic Irradiation: Investigations on a Possible Mechanism," *Radiat. Res.* *92*, 560.

Cushing, D.H. (1979), "The Monitoring of Biological Effects: The Separation of Natural Changes from Those Induced by Pollution," *Phil. Trans. R. Soc. Lond. B.* *286*, 597.

Daniel, G.E. and H.D. Park (1953), "Reproduction in Paramecium as Affected by Small Doses of X-Ray and Beta Radiation," *Proc. Soc. Exp. Biol.* *83*, 662.

Donaldson, L.R. and K. Bonham (1964), "Effects of Low-Level Chronic Irradiation of Chinook and Coho Salmon Eggs and Alevins," *Trans. Am. Fish. Soc.* *93*, 333.

Donaldson, L.R. and K. Bonham (1970), "Effects of Chronic Exposure of Chinook Salmon Eggs and Alevins to Gamma Irradiation," *Trans. Am. Fish. Soc.* *99*, 112.

Donaldson, L.R. and R.F. Foster (1957), "Effects of Radiation on Aquatic Organisms" in *The Effects of Atomic Radiation on Oceanography and Fisheries* Publ. 551 (National Academy of Sciences, Washington, DC), pp. 96-102.

Dunster, H.J., R.J. Garner, H. Howells, and L.F.U. Wix (1964), "Environmental Monitoring Associated with the Discharge of Low Activity Radioactive Waste from Windscale Works to the Irish Sea," Health Phys. 10, 353.

Dyer, R.S. (1976), "Environmental Surveys of Two Deepsea Radioactive Waste Disposal Sites Using Submersibles," in International Symposium on the Management of Radioactive Wastes from the Nuclear Fuel Cycle (International Atomic Energy Agency, Vienna), pp. 22-26.

Egami, N. (1969a), "Kinetics of Recovery from Injury after Whole-Body X-Irradiation of the Fish Oryzias latipes at Different Temperatures," Radiat. Res. 37, 192.

Egami, N. (1969b), "Temperature Effect on Protective Action by Cysteamine Against X-rays in the Fish, Oryzias latipes," Int. J. Radiat. Biol. 15, 393.

Egami, N. (1970), "Effects of Cysteamine Given Before and After X-irradiation Under Different Temperature Conditions on Mortality of the Fish, Oryzias latipes," Int. J. Radiat. Biol. 18, 391.

Egami, N. and H. Etoh (1966), "Effect of Temperature on the Rate of Recovery from Radiation-Induced Damage in the Fish, Oryzias latipes," Radiat. Res. 27, 630.

Egami, N. and A. Hama (1975), "Dose-Rate Effects on the Hatchability of Irradiated Embryos of the Fish, Oryzias latipes," J. Radiat. Biol. 28, 273.

Egami, N. and A. Hama (1977), "Effects of Chronic Gamma-Irradiation on the Fish, Oryzias latipes. I. Mortality of Fish and Hatchability of Eggs Laid by the Irradiated Fish," J. Radiat. Res. 18, 54.

Egami, N. and A. Hama (1979), "Effects of Chronic γ-Irradiation on the Fish, Oryzias latipes. V. Further Study of the Sterilized Fish Exposed to γ-rays During Development," J. Radiat. Res. 20, 15.

Egami, N. and A. Hama-Furukawa (1980), "Late Effects of Continuous γ-Irradiation of the Developmental Stage on the Gonads in *Oryzias latipes*," in *Radiation Effects on Aquatic Organisms*, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 105-117.

Egami, N. and A. Hama-Furukawa (1981), "Response to Continuous γ-Irradiation of Germ Cells in Embryos and Fry of the Fish, *Oryzias latipes*," *J. Radiat. Biol.* *40*, 563.

Egami, N. and Y. Hyodo-Taguchi (1971), "Species Difference in Radiosensitivity of Germ Cells Among Teleostean Fishes," *J. Radiat. Res.* *13*, 23.

Egami, N. and Y. Hyodo-Taguchi (1973), "Dominant Lethal Mutation Rates in the Fish *Oryzias latipes*, Irridiated at Various Stages of Gametogenesis," in *Proc. Symp. Genetics and Mutagenesis of Fish*, J.H. Schroeder, Ed. (Springer-Verlag, New York, NY), pp. 75-81.

Egami, N. and K.-I. Ijiri (1979), "Effects of Irradiation on Germ Cells and Embryonic Development in Teleosts," *Int. Rev. Cytol.* *59*, 195.

Egami, N., A. Shimada, and A. Hama-Furukawa (1983), "Dominant Lethal Mutation Rate After γ-Irradiation of the Fish, *Oryzias latipes*," *Mutat. Res.* *107*, 265.

Ejima, Y. and T. Shiroya (1982), "Photoreactivation Associated with Chromosomal Abonormality in Sea Urchin Eggs Fertilized with Ultraviolet-Irradiated Sperm," *Photochem. Photobiol.* 36, 37.

Emery, R.M. and D.C. Klopfer (1976), "The Distribution of Transuranic Elements in a Freshwater Pond Ecosystem," in *Environmental Toxicity of Aquatic Radionuclides: Models and Mechanisms*, W.W. Miller and J.N. Stannard, Eds. (Ann Arbor Science Publishers, Inc., Ann Arbor, MI), pp. 269-285.

Emery, R.M. and M.C. McShane (1980), "Nuclear Waste Ponds and Streams on the Hanford Site: An Ecological Search for Radiation Effects," *Health Phys.* *38*, 787.

Engel, D.W. (1967), "Effect of Single and Continuous Exposures of Gamma Radiation on the Survival and Growth of the Blue Crab, Callinectes sapidus," Radiat. Res. 32, 685.

Engel, D.W. (1979), "Experimental Approaches to Demonstrate the Interactions of Radiation with Other Environmental Stresses," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 301-316.

Engel, D.W., J.W. Angelovic, and E.M. Davis (1967), "Effects of Acute Irradiation on the Blood of Pinfish," in Ann. Rep. Bur. Comm. Fish. Radiobiol. Lab., Beaufort, NC, U.S. Fish Wildl. Serv. Circ. 270, pp. 35-38.

Engel, D.W., M. G.Shelton, and J.C. White, Jr. (1974), "The Effect of Ionizing Radiation and Salinity on the Grass Shrimp, Palaemonetes pugio," Radiat. Res. 59, 72.

Engel, D.W., J.C. White, Jr., and E.M Davis (1965), "Effect of Radiation on Marine Organisms," in Ann. Rep. Bur. Comm. Fish. Radiobiol. Lab., Beaufort, NC, U.S. Fish Wildl. Serv. Circ. 204, pp. 28-40.

Erickson, R.C. (1973), "Effects of Chronic Irradiation by Tritiated Water on Poecilia reticulata, the Guppy," in Proc. 3rd Natl. Symp. Radioecology, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1091-1099.

Etoh, H., and N. Egami (1967), "Damage Accumulation and Recovery in the Fish Oryzias latipes Exposed to Fractionated or Protracted Radiation at Different Temperatures," Radiat. Res. 32, 884.

Etoh, H., Y. Hyodo-Taguchi, and T. Maruyama (1968), "Effects of X-Irradiation of a Part of the Body on Mortality Rate and on Histological Changes in the Goldfish, Carassius auratus," J. Radiat. Res. 9, 141.

Etoh, H., Y. Hyodo-Taguchi, and T. Maruyama (1974), "Mortality Response of Goldfish after Exposure to Fast Neutrons," Radiat. Res. 59, 513.

Etoh, H., and I. Suyama (1980), "Effects of Radiation on Cultured Fish Cells," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 195-203.

Folsom, T.R. and T.M. Beasley (1973), "Contributions from the Alpha Emitter, Polonium-210, to the Natural Radiation Environment of the Marine Organisms," in Proc. Symp. Radioactive Contamination of the Marine Environment (International Atomic Energy Agency, Vienna), pp. 625-632.

Frank, M.L. (1973), "Sensitivity of Carp (Cyprinus carpio) Embryos to Acute Gamma Radiation," in Proc. 3rd Natl. Symp. Radioecology, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1106-1112.

Fujita, S. and N. Egami (1984), "Effect of Gamma Irradiation on the Reproductive System of the Pond Snail Physa acuta," Radiat. Res. 98, 362.

Gehrs, C.W., J.R. Trabalka, and E.A. Bardill (1975), "Sensitivity of Adult and Embryonic Calanoid Copepods to Acute Ionizing Radiation," Radiat. Res. 63, 382.

Gesell, T.F., G. De P. Burke, and K. Becker (1976), "An International Intercomparison of Environmental Dosimeters," Health Phys. 30, 125.

Ghoneum, M.M.H. and N. Egami (1980), "Effects of γ-Irradiation of Adult and Embryo of Oryzias latipes on Thymus Size," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 135-137.

Ghoneum, M.M.H., K. Ijiri, M.S. Hamed, O.M. Gabr, and N. Egami (1983), "Effects of Gamma-Rays on the Taste Buds of Embryos and Adults of the Fish Oryzias latipes," J. Radiat. Res. 24, 278.

Gorbman, A. (1963), "An Exploratory Study of Radiation Damage in the Thyroids of Coral Reef Fishes From the Eniwetok Atoll," in Proc. 1st Natl. Symp. Radioecology, V. Schultz and A.W. Klement, Eds. (Reinhold Publishing Corp., New York, NY), pp. 385-399.

Gray, J.S. (1979), "Pollution-Induced Changes in Populations," *Phil. Trans. R. Soc. Lond. B.* *286*, 545.

Green, E.L. (1968), "Genetic Effects of Radiation on Mammalian Populations," *Ann. Rev. Genet.* *2*, 87.

Grosch, D.S. (1962), "The Survival of *Artemia* Populations in Radioactive Sea Water," *Biol. Bull.* *123*, 302.

Grosch, D.S. (1966), "The Reproductive Capacity of *Artemia* Subjected to Successive Contaminations With Radiophosphorus," *Biol. Bull.* *131*, 261.

Guthrie, J.E. and R.A. Brust (1971), "Sterilization of *Aedes* *aegypti* (L.) as a Result of Chronic Exposure to Low Doses of Beta Radiation," *Nature* *232*, 345.

Guerrero, I., A. Villasante, V. Corces, A. Pellicer (1984), "Activation of a c-K-ras Oncogene by Somatic Mutation in Mouse Lymphomas Induced by Gamma Radiation," *Science* *225*, 1159.

Guthrie, J.E. and R.A. Brust (1973), "Effects of Chronic Radiation Exposure on Mosquitoes (Diptera: Culicidae). 1. Effects of Rearing in ^{90}Sr + ^{90}Y Solutions," in *Proc. 3rd Natl. Symp. Radioecology*, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1174-1178.

Hama, A. and N. Egami (1977), "Effect of Chronic γ-Irradiation on the Fish, *Oryzias* *latipes*. II. Dose-Rate Effect of γ-Rays on the Initiation of Mitosis in the Regeneration Area of the Tail Fin," *J. Radiat. Res.* *18*, 54.

Hama-Furukawa, A., and N. Egami (1980), "Effect of Irradiation on the Fin Regeneration of the Fish, *Oryzias* *latipes*, with Special Reference to the Dose Rate at Different Temperatures," in *Radiation Effects on Aquatic Organisms*, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 139-146.

Hamaguchi, S. (1980), "Differential Radiosensitivity of Germ Cells According to Their Developmental Stages in the Teleost, Orzias latipes," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 119-128.

Hansen, H.J.M. (1975), "Changes in the Lipid Fraction of Eel Gills after Ionizing Irradiation in Vivo and a Shift from Fresh to Sea Water," Radiat. Res. 62, 216.

Hansen, H.J.M. (1980), "Changes in the Lipid Fraction of Eel (Anguilla anguilla) Gills after Ionizing Irradiation and a Shift From Fresh to Sea Water: Further Influence of Temperature and Treatment with Actinomycin D," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 147-155.

Hansen, S.R., and R.R. Garton (1982), "Ability of Standard Toxicity Tests to Predict the Effects of the Insecticide Diflubenzuron on Laboratory Stream Communities," Can. J. Fish. Aquat. Sci. 39, 1273.

Harrison, F.L., D.W. Rice, Jr., D.H. Moore, and M. Varela (1985), "Effects of Radiation on Frequency of Chromosomal Aberrations and Sister Chromatid Exchange in the Benthic Worm Neanthes arenaceodentata," in Wastes in the Ocean: Biological Effects of Waste Disposal, Volume 9 (Robert E. Krieger, Malabar, FL).

Held, E.E. (1960), "Land Crabs and Fission Products at Eniwetok Atoll," Pac. Sci. 14, 18.

Hetherington, J.A., D.F. Jefferies, N.T. Mitchell, R.J. Pentreath, and D.S. Woodhead (1976), "Environmental and Public Health Consequences of the Controlled Disposal of Transuranic Elements to the Marine Environment," in Proc. Symp. Transuranium Nuclides in the Environment (International Atomic Energy Agency, Vienna), pp. 139-154.

Holton, R.L., C.L. Osterberg, and W.O. Forster (1973), "Effect of Gamma Irradiation on the Maintenance of Population Size in the Brine Shrimp, *Artemia*," in *Proc. 3rd Natl. Symp. Radioecology*, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1198-1205.

Holzberg, S. (1973), "Change of Aggressive Readiness in Post-Irradiation Generations of the Convict Cichlid Fish, *Cichlasoma nigrofasciatum*," in *Proc. Symp. Genetics Mutagenesis Fish*, J.H. Schroeder, Ed. (Springer-Verlag, New York, NY), pp. 173-176.

Holzberg, S. and J.H. Schroeder (1972), "Behavioural Mutagenesis in the Convict Cichlid Fish, *Cichlasoma nigrofasciatum* Guenther: I. The Reduction of Male Aggressiveness in the First Postirradiation Generation," *Mutat. Res.* *16*, 289.

Hoppenheit, M. (1973), "Effects of Fecundity and Fertility of Single Sub-Lethal X-Irradiation of *Gammarus duebeni* Females," in *Proc. Symp. Radioactive Contamination of the Marine Environment* (International Atomic Energy Agency, Vienna), pp. 479-486.

Horsley, R.J. and A. Laszlo (1973), "Additional Recovery in X-Irradiated *Oedogonium cardiacum* Can be Suppressed by Cyclohexaimide," *J. Radiat. Biol.* *23*, 201.

Hyodo, Y. (1964), "Effect of X-Irradiation on the Intestinal Epithelium of the Goldfish, *Carassius auratus*: I. Histological Changes in the Intestine of Irradiated Fish," *Annot. Zool. Jpn.* *37*, 104.

Hyodo, Y. (1965a), "Effect of X-Irradiation on the Intestinal Epithelium of the Goldfish, *Carassius auratus*: II. Influence of Temperature on the Development of Histopathological Changes in the Intestine," *Radiat. Res.* *24*, 133.

Hyodo, Y. (1965b), "Development of Intestinal Damage after X-Irradiation and H^3-Thymidine Incorporation into Intestinal Epithelial Cells of Irradiated Goldfish, *Carassius auratus*, at Different Temperatures," *Radiat. Res.* *26*, 383.

Hyodo-Taguchi, Y. (1970), "Effect of X-Irradiation on DNA Synthesis and Cell Proliferation in the Intestinal Epithelial Cells of Goldfish at Different Temperatures with Special Reference to Recovery Process," Radiat. Res. 41, 568.

Hyodo-Taguchi, Y. (1977), "Effects of Chronic Gamma Irradiation on the Spermatoganesis of the Fish, Oryzias latipes, with Special Reference to Exposure to Tritiated Water," J. Radiat. Res. 18, 55.

Hyodo-Taguchi, Y. (1980), "Effects of Chronic γ-Irradiation on Spermatogenesis in the Fish, Oryzias latipes, with Special Reference to Regeneration of Testicular Stem Cells," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 91-104.

Hyodo-Taguchi, Y. and N. Egami (1969), "Development of Intestinal Radiation Injury and Recovery at Different Temperatures in Fish," in Comparative Cellular and Species Radiosensitivity, V.P. Bond and T. Sugahara, Eds. (Williams and Wilkins Co., Baltimore, MD), pp. 244-254.

Hyodo-Taguchi, Y. and H. Etoh (1983), "The Fecundity and Fertility of Medaka Exposed to Chronic γ-Radiation in Their Embryonic Stages," J. Radiat. Res. 24, 270.

Hyodo-Taguchi, Y., H. Etoh, and N. Egami (1973), "RBE of Fast Neutrons for Inhibition of Hatchability in Fish Embryos Irradiated at Different Developmental Stages," Radiat. Res. 53, 385.

Ijiri, K-I. (1983), "Chromosomal Studies on Radiation-Induced Gynogenesis and Diploid Gynogenesis in the Fish Oryzias latipes," J. Radiat. Res. 24, 184.

Ishikawa, T., S. Takayama, and T. Kitagawa (1978), "Autoradiographic Demonstration of DNA Repair Synthesis in Ganglion Cells of Aquarium Fish at Various Age in vivo," Virchows Arch. B Cell Path. 28, 235.

Iwasaki, T. (1965), "Sensitivity of Artemia Eggs to γ-Irradiation: VII. Relationship Between the Degree of Biological Damage and the Decay of Free Radicals in Irradiated Eggs," Int. J. Radiat. Biol. 9, 573.

Johnson, T.S., R.E. Nakatani, and F.P. Conte (1970), "Influence of Temperature and X-Irradiation on the Cellular Dynamics of the Intestinal Epithelium in Coho Salmon Oncorhynchus kisutch," Radiat. Res. 42, 129.

Jordan, N.M. and D.J. Kimeldorf (1971), "The Visual Response of the Purple Shore Crab, Hemigrapsus nudus, to Ionizing and Non-Ionizing Radiations," Experientia 27, 653.

Joseph, A.B., P.F. Gustafson, I.R. Russel, E.A. Schuert, H.L. Volchok, and A. Tamplin (1971), "Sources of Radioactivity and their Characteristics," in Radioactivity in the Marine Environment (National Academy of Sciences, Washington, DC), pp. 6-41.

Kamath, P.R. (1969), "Recent Guidlines for Developing Environmental Monitoring Programs (Land and Water) Near Nuclear Installations," in Environmental Contamination by Radioactive Materials (International Atomic Energy Agency, Vienna), pp. 295-308.

Kaplan, H.S. and L.E. Moses (1964), "Biological Complexity and Radiosensitivity," Science 145, 21.

Kasatkina, S.V. (1973), "Effect of Strontium-90 + Yttrium-90 on the Development and Functioning of the Hatching Glands in Atlantic Salmon," in Effect of Ionizing Radiation on the Organism, Publ. AEC-tr-7418, B.P. Sorokin, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 66-74.

Kaufman, G.A. and R. J. Beyers (1973), "Effects of Chronic Gamma Irradiation on the Fish Oryzias latipes," in Proc. 3rd Natl. Symp. Radioecology, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1119-1124.

Keyl, H.G. (1958), "Untersuchungen am Karyotypus von Chironomus Thummi: II Strukturveraenderungen an den Speicheldrusen Chromosomen nach Roentgenbestrahlung von Embryonen und Larven," Chromosoma 9, 441.

Kimeldorf, D.J. and R. W. Fortner (1971), "The Prompt Detection of Ionizing Radiations by a Marine Coelenterate," Radiat. Res. 46, 52.

Kligerman, A.D. (1979), "Cytogenetic Methods for the Detection of Radiation-Induced Chromosome Damage in Aquatic Organisms," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems, Technical Report Series 190 (International Atomic Energy Agency, Vienna,), pp. 349-367.

Kligerman, A.D. (1980), "The Use of Aquatic Organisms to Detect Mutagens that Cause Cytogenetic Damage," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 241-252.

Kligerman, A.D., S.E. Bloom, and W.M. Howell (1975), "Umbra Limi: A Model for the Study of Chromosome Aberrations in Fishes," Mutat. Res. 31, 225.

Konno, K. (1980), "Effects of γ-Irradiation on the Gonads of the Rainbow Trout, Salmo gairdneri irideus, During Embryonic Stages," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 129-133.

Kooijman, S.A.L.M. and J.A.J. Metz (1984), "On the Dynamics of Chemically Stressed Populations: The Deduction of Population Consequences from Effects on Individuals," Ecotoxicol. Environ. Saf. 8, 254.

Kosheleva, V.V. (1973), "Change in Peripheral Blood in the Embryos and Larvae of Atlantic Salmon Under the Influence of Radioactive Contamination of the Water and X-Rays," in Effect of Ionizing Radiation on the Organism, Publ. AEC-tr-7418, B.P. Sorokin, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 75-88.

Krivolutsky, D., V. Turcaninova, and Z. Mikhaltsova (1982), "Earthworms as Bioindicators of Radioactive Soil Pollution," Pedobiologia 23, 263.

Krumholz, L.A. (1956), "Observations on the Fish Population of a Lake Contaminated by Radioactive Wastes," Am. Mus. Nat. Hist. Bull. 110, 281.

Kulikov, N.V. (1970), "Radiosensitivity of Tench (Tinca tinca L.) Embryos at Early Stages of Development," Radiobiology 10, 127 (AEC-tr-7109).

Lappenbusch, W. L., D. G. Watson, and W. L. Templeton (1971), "In Situ Measurement of Radiation Dose in the Columbia River," Health Phys. 21, 247.

Lasher, R. and R. Rugh (1962), "The Hertwig Effect in Teleost Development," Biol. Bull. 123, 582.

Leenhouts, H.P. and K.H. Chadwick (1978), "The Crucial Role of DNA Double-Strand Breaks in Cellular Radiobiological Effects," Adv. Radiat. Biol. 7, 55.

Lockner, D., A. Pettersson, and U. Ericson (1972), "Radiosensitivity of Haemopoiesis in Fish Studies at 18° and 10°C," J. Radiat. Biol. 21, 559.

Lomenick, T.F. and D.A. Gardiner (1965), "The Occurrence and Retention of Radionuclides in the Sediments of White Oak Lake," Health Phys. 11, 567.

Luckey, T.D. (1980), Hormesis with Ionizing Radiation (CRC Press, Inc., Boca Raton, FL), 222 p.

Mano, Y., K. Kator, and N. Egami (1982), "Photoreactivation and Excision Repair of Thymine Dimers in Ultraviolet-Irradiated Cultured Fish Cells," Radiat. Res. 90, 501.

Mano, Y., H. Mitani, H. Etoh, and N. Egami (1980), "Survival and Photoreactivability of Ultraviolet-Irradiated Cultured Fish Cells (CAF-MM1)," Radiat. Res. 84, 514.

Marine Protection, Research, and Sanctuaries Act (1972), 33 U.S.C. Secs. 1401 et seq. (U.S. GPO, Washington, DC).

Marshall, J.S. (1962), "The Effects of Continuous Gamma Radiation on the Intrinsic Rate of Natural Increase of Daphnia pulex," Ecology 43, 598.

Marshall, J.S. (1966), "Population Dynamics of Daphnia pulex as Modified by Chronic Radiation Stress," Ecology 47, 561.

Marshall, J.S. (1967), "Radiation Stress in Exploited Daphnia Populations, "Limnol..Oceanogr. 12, 154.

McCreery, M.J., C.E. Swenberg, M.J. Basso, J.J. Conklin, and J.-S. Hsieh Unpublished report, Biologic Dosimetry by Assessment of Radiation-Induced Free Radicals in Calcified Tissues.

McGregor, J.F. and H.B. Newcombe (1968), "Major Malformations in Trout Embryos Irradiated Prior to Active Organogenesis," Radiat. Res. 35, 282.

McGregor, J.F. and H.B Newcombe (1972a), "Dose-Response Relationships for Yields of Major Eye Malformations Following Low Doses of Radiation to Trout Sperm," Radiat. Res. 49, 155.

McGregor, J.F. and H.B. Newcombe (1972b), "Decreased Risk of Embryo Morality Following Low Doses of Radiation to Trout Sperm," Radiat. Res. 52, 536.

McNally, N.J. (1982), "Cell Survival," in Radiation Biology, D.J. Pizzarello, Ed. (CRC Press, Inc., Boca Raton, FL), pp. 28-68.

Metalli, P. (1979), "An Outline of Methodological Problems Related to the Assessment of Genetic Effects of Radiation," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 403-412.

Metalli, P. and E. Ballardin (1962), "First Results on X-Ray-Induced Genetic Damage in Artemia salina Leach," Atti. Assoc. Genet. Ital. 7, 219.

Metalli, P. and E. Ballardin (1972), "Radiobiology of Artemia: Radiation Effects and Ploidy," Curr. Top. Radiat. Res. Q. 7, 181.

Michibata, H. and N. Egami (1977), "Effect of Chronic Gamma-Irradiation on the Fish, Oryzia latipes. III. Effects on the Gonad with Special Reference to Spermatogonial Population," J. Radiat. Res. 18, 54.

Migalovskaya, V.N. (1973), "Chronic Effect of Strontium-90 + Yttrium-90 on the Frequency of Chromosomal Aberrations in the Embryonal Cells of the Atlantic Salmon," in Effect of Ionizing Radiation on the Organism, Publ. AEC-tr-7418, B.P. Sorokin, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 89-99.

Mitani, H. (1983), "Lethal and Mutagenic Effects of Radiation and Chemicals on Cultured Fish Cells Derived from the Erythrophoroma of Goldfish (Carassius auratus)," Mutat. Res. 107, 279.

Mitani, H. and N. Egami (1982), "Rejoining of DNA Strand Breaks after Gamma-Irradiation in Cultured Fish Cells, CAF-MM1," Int. J. Radiat. Biol. 44, 85.

Mitani, H., H. Etoh, and N. Egami (1982), "Resistance of a Cultured Fish Cell Line (CAF-MM1) to γ-Irradiation," Radiat. Res. 89, 334.

Mong, S.J. and T.M. Berra (1979), "The Effects of Increasing Dosages of X-Radiation on the Chromosomes of the Central Mudminnow, Umbra limi (Kirtland) (Salmoniformes: Umbridae)," J. Fish Biol. 14, 523.

Mount, D.I., N.A. Thomas, T.J. Norberg, M.T. Barbour, T.H. Roush, and W.F. Brandes (1984), Effluent and Ambient Toxicity Testing and Instream Community Response on the Ottawa River, Lima, Ohio, U.S. Environmental Protection Agency, Washington, DC, EPA-600/3-84-080.

National Advisory Committee on Oceans and Atmosphere (1984), Nuclear Waste Management and the Use of the Sea, draft report (Washington, DC).

National Research Council, Committee on the Biological Effects of Ionizing Radiations (1980), The Effects on Populations of Exposure to Low Levels of Ionizing Radiation: 1980 (National Academy Press, Washington, DC).

Neuhold, J.M. and R.K. Sharma (1969), "Interacting Effects of Gamma Radiation and Sodium Halide Concentrations on Rainbow Trout," in Proc. 2nd Natl. Symp. Radioecology, CONF-670503, D.J. Nelson and F.C. Evans, Eds. (U.S. Atomic Energy Commission, Washington, DC), pp. 142-151.

Newcombe, H.B. (1971), "The Genetic Effects of Ionizing Radiations," Adv. Genet. 16, 239.

Newcombe, H.B., and J.F. McGregor (1967), "Major Congenital Malformations from Irradiations of Sperm and Eggs," Mutat. Res. 4, 663.

Newcombe, H.B., and J.F. McGregor (1972), "Increased Embryo Production Following Low Doses of Radiation to Trout Spermatozoa," Radiat. Res. 51, 402.

Noshkin, V.E., K.M. Wong, T.A. Jokela, R.J. Eagle, and J.L. Brunk (1978), Radionuclides in the Marine Environment near the Farallon Islands, Lawrence Livermore National Laboratory, Livermore, CA, UCRL-52381.

OECD/NEA (1980), Review of the Continued Suitability of the Dumping Site for Radioactive Waste in the North-East Atlantic, Nuclear Energy Agency, Organisation for Economic Cooperation and Development (OECD/NEA), Paris, France.

OECD/NEA (1985), Review of the Continued Suitability of the Dumping Site for Radioactive Waste in the Northeast Atlantic, Nuclear Energy Agency, Organisation for Economic Cooperation and Development (OECD/NEA), Paris, France.

Oganesyan, S.A. (1973), "Histogenesis and Functioning of the Hypophysis and Thyroid Gland in the Larvae of Alantic Salmon Exposed to Ionizing Radiation," in Proc. 3rd Natl. Symp. Radioecology, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 53-65.

Ophel, I.L. (1976), "Effects of Ionizing Radiation on Aquatic Organisms," in Effects of Ionizing Radiation on Aquatic Organisms and Ecosystems, Technical Report Series 172 (International Atomic Energy Agency, Vienna), pp. 57-88.

Ophel, I.L. (1979), "Radiation Effects in Contaminated Aquatic Ecosystems and Their Relation to Laboratory Studies," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 211-216.

Pantelias, G.E. and H.D. Maillie (1983), "A Simple Method for Premature Chromosome Condensation Induction in Primary Human and Rodent Cells Using Polyethylene Glycol," Somatic Cell Genet. 9, 533.

Patel, B. (1975), "Field and Laboratory Comparability of Radioecological Studies," in Design of Radiotracer Experiments in Marine Biological Systems, Technical Report Series 167 (International Atomic Energy Agency, Vienna), pp. 211-240.

Patel, B. and S. Patel (1979), "Techniques for Detecting Radiation Effects on Biochemical and Physiological Systems in Aquatic Organisms," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 237-265.

Perkins, E. J. (1979), "The Need for Sublethal Studies," Phil. Trans. R. Soc. Lond. B. 286, 425.

Pohl Ruling, J. and P. Fischer (1979), "The Dose Effect Relationship of Chromosome Aberrations to Alpha and Gamma Irradiation in a Population Subjected to an Increased Burden of Natural Radioactivity," Rad. Res. 80, 61.

Polikarpov, G.G. (1966), Radioecology of Aquatic Organisms (Reinhold, New York, NY).

Polikarpov, G.G. (1979), "Experimental Methods for Radiobiological Investigations with Developing Fish Eggs," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 173-194.

Preston, A. (1959), "Leucocytosis in Response to Bacteremia as a Feature of the Acute Radiation Syndrome in the Plaice," Nature 183, 832.

Preston, A. (1969), "Aquatic Monitoring Programmes," in Environmental Contamination by Radioactive Materials (International Atomic Energy Agency, Vienna), pp. 309-324.

Preston, A., and D.F. Jefferies (1969), "Aquatic Aspects in Chronic and Acute Contamination Situations," in Environmental Contamination by Radioactive Materials (International Atomic Energy Agency, Vienna), pp. 183-210.

Purdom, C.E. (1966), "Radiation and Mutation in Fish," in Proc. Symp. Disposal of Radioactive Wastes into Seas, Oceans, and Surface Waters (International Atomic Energy Agency, Vienna), pp. 861-867.

Purdom, C.E. and D.S. Woodhead (1973), "Radiation Damage in Fish," in Proc. Symp. Genetics and Mutagenesis of Fish, J.H. Schroeder, Ed. (Springer-Verlag, New York, NY), pp. 67-73.

Ravera, O. (1966), "Effects of X-Irradiation on Various Stages of the Life Cycle of Physa acuta, Draparnaud, a Fresh-Water Gastropod," in Proc. Symp. Disposal of Radioactive Wastes into Seas, Oceans, and Surface Waters (International Atomic Energy Agency, Vienna), pp. 799-808.

Ravera, O. (1967), "The Effect of X-Rays on the Demographic Characteristics of Physa acuta (Gastropoda: Basommatophora)," Malacologia 5, 95.

Regan, J.D. and J.S. Cook (1967), "Photoreactivation in an Estabalished Vertebrate Cell Line," Proc. Nat. Acad. Sci. USA 58, 2274.

Regan, J.D., R.D. Snyder, A.A. Francis, and B.L. Olla (1983), "Excision Repair of Ultraviolet- and Chemically-Induced Damage in the DNA of Fibroblasts Derived from two Closely Related Species of Marine Fishes," *Aquat. Toxicol.* *4*, 181.

Rice, T.R. and J.P. Baptist (1970), "Ecological Aspects of Radioactivity in the Marine Environment," in *Proc. Symp. Environmental Radioactivity*, J.C. Clapton, Ed. (Department of Geography and Environmental Engineering, Johns Hopkins University, Baltimore, MD), pp. 107-180.

Roop, R.D. and C.T. Hunsaker (1985), "Biomonitoring for Toxics Control in NPDES Permitting," *J. Water Pollut. Control Fed.* *57*, 271.

Rugh, R. and H. Clugston (1955), "Effects of Various Levels of X-Irradiation on the Gametes and Early Embryos of *Fundulus heteroclitus*," *Biol. Bull.* *108*, 318 .

Scarborough, B.B. and R.G. Addison (1962), "Conditioning in Fish: Effects of X-Irradiation," *Science* *136*, 712.

Schendel, P.F. (1981), "Inducible Repair Systems and Their Implications for Toxicology," *CRC Critical Reviews in Toxicology* *8*, 311.

Schroeder, J.H. (1969a), "Inheritance of Radiation-Induced Spinal Curvatures in the Guppy *Lebistes reticulatus*," *Can. J. Genet. Cytol.* *11*, 937.

Schroeder, J.H. (1969b), "X-Ray-Induced Mutations in the Poeciliid Fish, *Lebistes Reticulatus* Peters," *Mutat. Res.* *7*, 75.

Schroeder, J.H. (1969c), "Radiation-Induced Spermatogonial Exchange Between the X and Y Chromosomes in the Guppy," *Can. J. Genet. Cytol.* *11*, 948.

Schroeder, J.H. (1979), "Methods for Screening Radiation-Induced Mutations in Fish," in *Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems*, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 381-402.

Schroeder, J.H. (1980), "Radiation-Induced Mutations in Fish," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 217-222.

Schroeder, J.H. and S. Holzberg (1972), "Population Genetics of Lebistes (Poecilia) Reticulatus Peters (Poeciliidae; Pisces). I. Effects of Radiation-Induced Mutations on the Segregation Ratio In Postirradiation F_2," Genetics 70, 621.

Setlow, R.B. (1982), "Dose Response Relations: The Effects of DNA Repair," in Proc. Symp. Genetic Toxicology: An Agricultural Perspective, R.A. Flech and A. Hollaender, Eds. (Plenum Press, New York, NY), pp. 391-401.

Shechmeister, I.L., L.J. Watson, V.W. Cole, and L.L. Jackson (1962), "The Effect of X-Irradiation of Goldfish: Effect of X-Irradiation on Survival and Susceptibility of the Goldfish, Carassius auratus, to Infection by Aeromonas salmonicida and Gyrodactylus spp.," Radiat. Res. 16, 89.

Slobodkin, L.B. and S. Richman (1956), "The Effect of Removal of Fixed Percentages of the Newborn on Size and Variability in Populations of Daphnia pulicaria (Forbes)," Limnol. Oceanogr. 1, 209.

Squire, R.D. (1970), "The Effects of Acute Gamma Irradiation on the Brine Shrimp, Artemia. II. Female Reproductive Performance," Biol. Bull. 139, 375.

Squire, R.D. (1973), "The Effects of Acute Gamma Irradiation on the Brine Shrimp, Artemia. III. Male F_1 Reproductive Performance Following Paternal Irradiation of Mature Sperm," Biol. Bull. 144, 192.

Strand, J.A., M.P. Fujihara, W.L. Templeton, and E.G. Tangen (1973a), "Suppression of Chondrococcus columnaris Immune Response in Rainbow Trout Sub-Lethally Exposed to Tritiated Water During Embryogenesis," in Proc. Symp. Radioactive Contamination of the Marine Environment (International Atomic Energy Agency, Vienna), pp. 543-550.

Strand, J.A., W.L. Templeton, and E.G. Tangen (1973b), "Accumulation and Retention of Tritium (Tritiated Water) in Embryonic and Larval Fish, and Radiation Effect," in Proc. 3rd Natl. Symp. Radioecology, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 445-451.

Styron, C.E. (1971), "Effects of Beta and Gamma Radiation on a Population of Springtails, Sinella curviseta (Collembola)," Radiat. Res. 48, 53.

Suyama, I., H. Etoh, and R. Ichikawa (1980), "Effects of Ionizing Radiation on the Development of Limanda Eggs," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 205-207.

Suyama, I. and H. Etoh (1983), "X-Ray-Induced Dicentric Yields in Lymphocytes of the Teleost, Umbra limi," Mutat. Res. 107, 111.

Suzuki, J. and N. Egami (1983), "Mortality of the Earthworms, Eisenia foetida, after γ-Irradiation at Different Stages of Their Life History," J. Radiat. Res. 24, 209.

Szechter, A. and G. Schwarz (1977), "Dose-Rate Effects, Fractionation, and Cell Survival at Lowered Temperatures", Radiat. Res. 71, 593.

Templeton, W.L. (1966), "Resistance of Fish Eggs to Acute and Chronic Irradiation," in Proc. Symp. Disposal of Radioactive Wastes into Seas, Oceans, and Surface Waters (International Atomic Energy Agency, Vienna), pp. 847-860.

Templeton, W.L. (1976a), "Effects of Ionizing Radiation on Aquatic Populations and Ecosystems," Technical Report Series 172 (International Atomic Energy Agency, Vienna), pp. 89-119.

Templeton, W.L. (1976b), "Effects of Radiation on Aquatic Populations," in Environmental Toxicity of Aquatic Radionuclides: Models and Mechanisms, W.W. Miller and J.N. Stannard, Eds. (Ann Arbor Science Publishers, Inc., Ann Arbor, MI), pp. 287-295.

Templeton, W.L. (1980), "Ecological Effects of Transuranics in the Marine Environment," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 3-12.

Templeton, W.L., R.E. Nakatani, and E.E. Held (1971), "Radiation Effects," in Radioactivity in the Marine Environment (National Academy of Sciences, Washington, DC), pp. 223-239.

Till, J.E., S.V. Kaye, and J.R. Trabalka (1976), The Toxictity of Uranium and Plutonium to the Developing Embryos of Fish, Oak Ridge National Laboratory, Oak Ridge, TN, ORNL-5160.

Trabalka, J.R. and C. P. Allen (1977), "Aspects of Fitness of a Mosquitofish Gambusia affinis Population Exposed to Chronic Low-Level Environmental Radiation," Radiat. Res. 70, 198.

Tsytsugina, V.G. (1973), "Effect of Incorporated Radionuclides on the Chromosomal Apparatus of Marine Fish," in Effect of Ionizing Radiation on the Organism, Publ. AEC-tr-7418, B.P. Sorokin, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 157-165.

Tsytsugina, V.G. (1979), "Methods of Demonstrating Cytogenetic Radiation Damage to Aqueous Organisms in vivo or in Systems of Tissue Cultures," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 369-380.

Turner, F.B., P. Licht, J.D. Thrasher, P.A. Medica, and J.R. Lannom, Jr. (1973), "Radiation-Induced Sterility in Natural Populations of Lizards (Crotaphytus wislizenii and Cnemidorus tigris)," in Radionuclides in Ecosystems (National Technical Information Service, Springfield, VA), pp. 1131-1143.

Tuschl, H., R. Kovac, and H. Altmann (1983), "UDS and SCE in Lymphocytes of Persons Occupationally Exposed to Low Levels of Ionizing Radiation," Health Phys. 45, 1.

Ulrikson, G.U. (1973), "Radiation Effects on Serum Proteins, Hematocrits, Electrophoretic Patterns and Protein Components in the Bluegill (Lepomis macrochirus)," in Proc. 3rd Natl. Symp. Radioecology, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1100-1105.

United Nations Scientific Committee on the Effects of Atomic Radiation (1977), Sources and Effects of Ionizing Radiation, E.77.IX.1 (United Nations Publications, New York, NY).

United Nations Scientific Committee on the Effects of Atomic Radiation (1982), Ionizing Radiation: Sources and Biological Effects, E.82.IX.8 (United Nations Publications, New York, NY).

Upton, A.C., A.W. Kimball, J. Furth, K.W. Christenberry, and W.H. Benedict (1960), "Some Delayed Effects of Atom-Bomb Radiations in Mice," Cancer Res. 20, 3.

Van Rijn, J., J. Van Den Berg, J.B.A. Kipp, D.H.J. Schamhart, and R. Van Wijk (1985), "Effect of Hypothermia on Cell Kinetics and Response to Hyperthermia and X Rays," Radiat. Res. 101, 292.

Wadley, G.W. and A. D. Welander (1971), "X-Rays and Temperature: Combined Effects on Mortality and Growth of Salmon Embryos," Trans. Am. Fish. Soc. 100, 267.

Walden, S.J. (1973), "Effects of Tritiated Water on the Embryonic Development of the Three-Spine Stickleback, Gasterosteus aculeatus Linnaeus," in Proc. 3rd Natl. Symp. Radionuclides in Ecosystems, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1087-1090.

Walton, D.G., A.B. Acton, and H.F. Stich (1983), "DNA Repair Synthesis in Cultured Mammalian and Fish Cells Following Exposure to Chemical Mutagens," Mutat. Res. 124, 153.

Ward, E., S.A. Beach, and E.D. Dyson (1971), "The Effect of Acute X-Irradiation on the Development of the Plaice, Pleuronectes platessa L.," J. Fish Biol. 3, 251.

Watson, D.G. and W.L. Templeton (1973), "Thermoluminescent Dosimetry of Aquatic Organisms," in Proc. 3rd Natl. Symp. Radionuclides in Ecosystems, CONF-710501, D.J. Nelson, Ed. (U.S. Atomic Energy Commission, Washington, DC), pp. 1125-1130.

Welander, A.D. (1954), "Some Effects of X-Irradiation of Different Embryonic Stages of the Trout (Salmo gairdnerii)," Growth 18, 227.

Welander, A.D., L.R. Donaldson, R.F. Foster, K. Bonham, and A.H. Seymour (1948), "The Effects of Roentgen Rays on the Embryos and Larvae of the Chinook Salmon," Growth 12, 203.

Whicker, F.W. (1980), "Ecological Effects of Transuranics in the Terrestrial Environment," in Transuranic Elements in the Environment, W.C. Hansen, Ed. (Office of Scientific and Technical Information Center, U.S. Department of Energy, Oak Ridge, TN), pp. 701-713.

Whicker, F.W., and V. Schultz (1982), Radioecology: Nuclear Energy and the Environment: Vol. II (CRC Press, Boca Raton, FL).

White, Jr., J.C., J.W. Angelovic, D.W. Engel, and E.M. Davis (1967), "Interactions of Radiation, Salinity, and Temperature on Estuarine Organisms," in Ann. Rep. Bur. Comm. Fish. Radiobiol. Lab., Circ. 270 (U.S. Fish Wildl. Serv., Beaufort, NC), pp. 29-35.

Willis, D.L. (1980), "The Effect of Temperature on the Radiation Response of the Rough-Skinned Newts, Taricha granulosa," in Radiation Effects on Aquatic Organisms, N. Egami, Ed. (Japan Scientific Societies Press, Tokyo; University Park Press, Baltimore, MD), pp. 157-167.

Willis, D.L. and W.L. Lappenbusch (1976), "The Radiosensitivity of the Rough-Skinned Newt (Taricha granulosa)," in Proc. 4th Natl. Symp. Radioecology, C.E. Cushing, Jr., Ed. (Dowden, Hutchinson, & Ross, Stroudsburg, PA), pp. 363-375.

Wolf, K. and J.A. Mann (1980), "Poikilotherm Vertebrate Cell Lines and Viruses: A Current Listing for Fishes," In Vitro 16, 16B.

Wolff, S. (1968), "Chromosome Aberrations and the Cell Cycle," Radiat. Res. 33, 609.

Woodhead, A.D., P. Achey, R.B. Setlow, and E. Grist (1978), "Photoenzymatic Repair of Ultraviolet-Irradiated DNA in the Cells of a Shark, Prionace glauca," Comp. Biochem. Physiol. 60B, 205.

Woodhead, A.D. and P.M. Achey (1979), "Photoreactivating Enzyme in the Blind Cave Fish, Anoptichthys jordani," Comp. Biochem. Physiol. 63B, 73.

Woodhead, A.D., R.B. Setlow, and R.W. Hart (1979), "Genetically Uniform Strains of Fish as Laboratory Models for Experimental Studies of the Effects of Ionizing Radiation," in Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 317-333.

Woodhead, D.S. (1970), "The Assessment of the Radiation Dose to Developing Fish Embryos Due to the Accumulation of Radioactivity by the Egg," Radiat. Res. 43, 582.

Woodhead, D.S. (1973a), "Levels of Radioactivity in the Marine Environment and the Dose Commitment to Marine Organisms," in Proc. Symp. Radioactive Contamination of the Marine Environment (International Atomic Energy Agency, Vienna), pp. 499-525.

Woodhead, D.S. (1973b), "The Radiation Dose Received by Plaice (Pleuronectes platessa) from the Waste Discharged into the North-East Irish Sea from the Fuel Reprocessing Plant at Windscale," Health Phys. 25, 115.

Woodhead, D.S. (1976), "Influence of Acute Irradiation on Induction of Chromosome Aberrations in Cultured Cells of the Fish *Ameca splendens*," in *Proc. Symp. Biol. Environ. Eff. of Low-Level Radiat.*, Vol. I (International Atomic Energy Agency, Vienna), pp. 67-76.

Woodhead, D.S. (1977), "The Effects of Chronic Irradiation on the Breeding Performance of the Guppy, *Poecilia reticulata* (Osteichthyes: Teleostei)," *Int. J. Radiat. Biol.* *32*, 1.

Woodhead, D.S. (1979), "Methods of Dosimetry for Aquatic Organisms," in *Methodology for Assessing Impacts of Radioactivity on Aquatic Ecosystems*, Technical Report Series 190 (International Atomic Energy Agency, Vienna), pp. 43-96.

Woodhead, D.S. and R.J. Pentreath (1983), "A Provisional Assessment of Radiation Regimes in Deep Ocean Environments," in *Wastes in the Oceans*, Vol. 3 (John Wiley, New York, NY), pp. 133-152.

Woodhead. D.S., C.J. Barker, and B.D. Rackham (1983), "The Effects of Chronic γ-Irradiation on Experimental Fish Populations: A Preliminary Account", Abstract in *Biological Effects of Low-level Radiation* (International Atomic Energy Agency, Vienna), pp. 646-648.

Woodhead, D.W. (1984), "Contamination Due to Radioactive Materials," in *Marine Ecology*, Vol. V, Part 3, O. Kinne, Ed. (John Wiley and Sons, Ltd., Chichester, UK), 1618 pp.

APPENDIX I

List of Major Review Articles

Blaylock, B.G. and J.R. Trabalka (1978), "Evaluating the Effects of Ionizing Radiation on Aquatic Organisms," in Advances in Radiation Biology, Vol. 7, J.T. Lett and H. Adler, Eds. (Academic Press, New York, NY), pp. 103-152.

Chipman, W.A. (1972), "Ionizing Radiation," in Marine Ecology, Vol. I, Pt. 3, Ch. 11, O. Kinne, Ed. (John Wiley/Interscience, New York, NY), pp. 1579-1657.

Egami, N. and K.I. Ijiri (1979), "Effects of Irradiation on Germ Cells and Embryonic Development in Teleosts," Int. Rev. Cytol. 59, 195.

Ophel, I.L. (1976), "Effects of Ionizing Radiation on Aquatic Organisms," in Effects of Ionizing Radiation on Aquatic Organisms and Ecosystems, Technical Report Series 172 (International Atomic Energy Agency, Vienna), pp. 57-88.

Polikarpov, G.G. (1966), Radioecology of Aquatic Organisms (Reinhold, New York, NY).

Templeton, W.L. (1976), "Effects of Ionizing Radation on Aquatic Populations and Ecosystems," Technical Report Series 172 (International Atomic Energy Agency, Vienna), pp. 89-119.

Templeton, W.L., R.E. Nakatani, and E.E. Held (1971), "Radiation Effects," in Radioactivity in the Marine Environment, (National Academy of Sciences, Washington, DC), 272.

Woodhead, D.W. (1984), Contamination Due to Radioactive Materials," in Marine Ecology, Vol. V, Part 3, O. Kinne, Ed. (John Wiley and Sons, Ltd., Chichester, UK), 1618.

APPENDIX II

Reference Summary Table

To conduct a systematic critical examination of the literature, we first classified articles according to effect endpoints and examined modifying factors. After reviewing the nearly 200 articles presented in this summary table, we selected only the most pertinent references for inclusion in the text and tables. However, all papers are given in the list of references. We then added further articles to the text and reference list, but did not add them to this summary table.

Appendix II. Reference summary table

		General sorts					Effect endpoints												Modifying factors								
Author	Year	Fish	Inverte-brates	Other	Acute	Chronic	Morta-lity	Pathophysiology	Repro-duction	Develop-ment	Growth	Life span	Meta-bolism	Popu-lation	Geno-toxicity	Beha-vioral	Cell culture	Other	Dose rate	RBE	Life stage	Phylogeny	Temperature	Salinity	Oxygen	Cell cycling	DNA repair
Abrahamson, S. (+)	73			X	X										X												
Anders, A. (+)	73a	X			X										X												
Anders, A. (+)	73b	X			X										X												
Angelovic, J.W. (+)	67	X			X		X																X	X			
Angelovic, J.W. (DWE)	68		X		X			X					X											X			
Aoki, K. (+)	66	X			X		X	X																			
Ayala, F.J.	66		X		X									X	X												
Bacq, Z.M. (PA)	61	X	X	X			X																				
Blaylock, B.G.	66		X			X								X	X												
Blaylock, B.G.	66		X			X								X	X												
Blaylock, B.G.	69	X				X			X	X				X													
Blaylock, B.G. (PGK)	69		X		X										X												
Blaylock, B.G. (TJM)	69	X			X		X																X				
Blaylock, B.G. (NAG)	71	X			X		X			X																	
Blaylock, B.G. (HHS)	72		X		X									X	X												
Blaylock, B.G.	73		X			X									X												
Blaylock, B.G. (JPW)	75					X												X			X						
Blaylock, B.G. (MLF)	80	X				X			X	X				X													
Bonham, K. (ADW)	63	X			X		X														X						
Bonham, K. (LRD)	66	X				X	X		X	X	X																
Bonham, K. (LRD)	72	X				X			X										X								
Brown, F.A.	63		X													X											
Brown, F.A. (YHP)	64		X			X										X											
Brown, V.M. (WLT)	64	X				X	X			X																	
Cabela, E. (+)	72			X	X																					X	X
Calkins, J.	64		X		X		X																				
Chee, P.C. (+)	79	X				X	X												X								
Chipman, W.A.	72	X	X		X	X	X	X	X	X	X	X	X	X	X	X		X									
Conte, F.P.	65	X			X		X	X																X			
Cooley, J.L. (FLM)	71		X			X	X		X										X								
Cooley, J.L.	73		X			X			X																		

Appendix II. Reference summary table

Author	Year	General sorts					Effect endpoints												Modifying factors								
		Fish	Invertebrates	Other	Acute	Chronic	Mortality	Pathophysiology	Reproduction	Development	Growth	Life span	Metabolism	Population	Genotoxicity	Behavioral	Cell culture	Other	Dose rate	RBE	Life stage	Phylogeny	Temperature	Salinity	Oxygen	Cell cycling	DNA repair
Cosgrove, G.E. (BGB)	73	X			X	X	X	X															X				
Cosgrove, G.E. (+)	75	X			X	X		X																			
Crenshaw, J.W.	65		X		X										X												
Croute, F. (+)	82		X			X					X																
Daniel, G.E. (HDP)	53		X						X										X								
Donaldson, L.R. (KB)	64	X				X	X		X	X	X			X					X								
Donaldson, L.R. (KB)	70	X				X			X		X			X					X								
Dunster, H.J. (+)	64																	X									
Egami, N. (HE)	66	X			X		X												X				X				
Egami, N.	69	X					X	X																		X	
Egami, N.	69	X			X		X											X					X				
Egami, N.	70	X			X		X											X					X				
Egami, N. (AH)	75	X			X	X	X		X						X				X			X		X			
Egami, N. (AH)	77	X			X		X												X								
Egami, N. (AH)	79	X				X		X	X		X								X								
Egami, N. (K-II)	79	X			X	X	X		X	X									X	X	X					X	X
Egami, N. (AH-F)	80	X				X	X	X	X										X		X						
Egami, N. (AH-F)	81	X				X	X		X										X								
Egami, N. (+)	83	X			X		X	X							X				X								
Egami, N. (Y H-T)	71	X				X		X	X																		
Egami, N. (YH-T)	73	X			X				X						X												
Emergy, R.M. (MCM)	80	X	X	X		X								X													
Emery, R.M. (DCK)	76	X	X	X		X												X									
Engel, D.W. (+)	65	X			X		X	X	X														X	X			
Engel, D.W. (+)	67	X			X			X																			
Engel, D.W.	67		X		X	X	X				X																
Engel, D.W. (+)	74		X		X			X																	X		
Engel, D.W.	79	X	X		X	X	X											X					X	X			
Erickson, R.C.	73	X				X	X			X	X					X			X		X		X				

Appendix II. Reference summary table

		General sorts					Effect endpoints												Modifying factors								
Author	Year	Fish	Invertebrates	Other	Acute	Chronic	Mortality	Pathophysiology	Reproduction	Development	Growth	Life span	Metabolism	Population	Genotoxicity	Behavioral	Cell culture	Other	Dose rate	RBE	Life stage	Phylogeny	Temperature	Salinity	Oxygen	Cell cycling	DNA repair
Etoh, H. (NE)	67	X			X	X	X												X				X				
Etoh, H. (+)	68	X			X		X	X																			
Etoh, H. (+)	74	X			X		X	X												X							
Etoh, H. (IS)	80	X			X		X										X		X								X
Folsom, T.R. (TMB)	73	X	X			X												X									
Frank, M.L.	73	X			X		X			X											X						
Fujita, S. (NE)	84		X		X		X		X	X																	
Gehrs, C.W. (+)	75		X		X		X			X											X						
Gesell, T.F. (+)	76																	X									
Ghoneum, M.M.H. (+)	83	X			X			X																			
Ghoneum, M.M.H. (NE)	80	X			X			X													X						
Gorbman, A. (MSJ)	63	X				X		X																			
Green, E.L.	68			X	X	X									X												
Grosch, D.S.	62		X			X			X			X		X													
Grosch, D.S.	66		X			X	X		X			X															
Guthrie, J.E. (RAB)	71		X	X		X			X																		
Guthrie, J.G. (RAB)	73		X			X			X							X			X		X						
Hama, A. (NE)	77	X				X			X										X								
Hama-Furukawa, A. (NE)	80	X			X						X							X	X					X		X	
Hamaguchi, S.	80	X			X				X																	X	
Hansen, H.J.M.	75	X			X			X																	X		
Hansen, H.J.M.	80	X			X			X															X	X			
Held, E.E.	60		X			X												X									
Hetherington, J.A. (+)	76	X	X	X		X												X	X								
Holton, R.L. (+)	73		X		X				X					X													
Holzberg, S. (JHS)	72	X			X										X	X											
Holzberg, S.	73	X			X										X												
Hoppenheit, M.	73		X		X		X		X		X																
Horsley, R.J. (AL)	73			X		X											X		X							X	
Hyodo, Y.	64	X			X			X																			
Hyodo, Y.	65a	X			X		X	X															X				
Hyodo, Y.	65b	X			X			X															X				

Appendix II. Reference summary table

		General sorts					Effect endpoints												Modifying factors								
Author	Year	Fish	Invertebrates	Other	Acute	Chronic	Mortality	Pathophysiology	Reproduction	Development	Growth	Life span	Metabolism	Population	Genotoxicity	Behavioral	Cell culture	Other	Dose rate	RBE	Life stage	Phylogeny	Temperature	Salinity	Oxygen	Cell cycling	DNA repair
Hyodo-Taguchi, Y. (NE)	69	X			X		X	X																X			
Hyodo-Taguchi, Y.	70	X			X			X															X			X	
Hyodo-Taguchi, Y. (+)	73	X			X		X			X										X		X					
Hyodo-Taguchi, Y.	77	X				X			X										X	X							
Hyodo-Taguchi, Y.	80	X				X			X										X							X	
Hyodo-Taguchi, Y. (HE)	83	X			X	X	X		X	X									X								
Ijiri, K.-I.	83	X			X										X												
Iwasaki, T.	65		X		X			X		X																	
Johnson, T.S. (+)	70	X			X			X															X			X	
Jordan, N.M. (DJK)	71		X															X									
Kamath, P.R.	69			X		X												X									
Kaplan, H.S. (LEM)	64			X	X													X									
Kasatkina, S.V.	73	X				X		X																			
Kaufman, G.A. (RJB)	73	X				X		X																			
Keyl, H.G.	58		X		X										X												
Kimeldorf, D.J. (RWF)	71		X		X											X			X								
Kligerman, A.D.	79	X													X												
Kligerman, A.D.	80	X	X			X									X												
Konno, K.	80	X			X		X		X																		
Kosheleva, V.V.	73	X				X		X		X																	
Krivolutsky, D. (+)	82			X		X								X													
Krumholz, L.A.	56	X				X					X			X				X									
Kulikov, N.V.	70	X			X		X			X											X						
Lappenbusch, W.L. (+)	71			X		X													X								
Lasher, R. (RR)	62	X			X					X																	
Leenhouts, H.P. (KHC)	78				X	X									X				X	X							X
Lockner, D. (+)	72	X			X		X	X																X			
Lomenick, T.F. (DAG)	65																	X									
Marshall, J.S.	62		X			X			X			X		X													
Marshall, J.S.	66		X			X								X													
Marshall, J.S.	67		X			X								X					X								

Appendix II. Reference summary table

		General sorts					Effect endpoints												Modifying factors								
Author	Year	Fish	Invertebrates	Other	Acute	Chronic	Mortality	Pathophysiology	Reproduction	Development	Growth	Life span	Metabolism	Population	Genotoxicity	Behavioral	Cell culture	Other	Dose rate	RBE	Life stage	Phylogeny	Temperature	Salinity	Oxygen	Cell cycling	DNA repair
McGregor, J.F. (HBN)	68	X			X		X			X											X						
McGregor, J.F. (HBN)	72	X			X										X												
McGregor, J.F. (HBN)	72	X			X		X			X																	
Metalli, P.	79														X				X								
Michibata, H. (NE)	77	X				X			X																		
Migalovskaya, V.N.	73	X			X	X									X												
Neuhold, J.M. (RKS)	67	X			X		X											X									
Newcombe, H.B. (JFM)	67	X			X		X			X																	
Newcombe, H.B.	71	X	X	X	X	X									X				X								
Newcombe, H.B. (JFM)	72	X			X				X																		
Oganesyan, S.A.	73	X			X	X		X																			
Ophel, I.L.	79	X	X			X												X	X								
Patel, B.	75			X														X					X	X			
Patel, B. (S.P.)	79	X	X					X					X					X									
Polikarpov, G.G.	79	X								X								X									
Preston, A.	59	X			X			X																			
Preston, A.	69					X												X									
Preston, A. (DFJ)	69	X	X	X	X	X												X									
Purdom, C.E.	66	X			X										X												
Purdom, C.E. (DSW)	73	X			X	X			X						X				X								
Ravera, O.	66		X		X		X		X												X						
Ravera, O.	67		X		X		X		X										X								
Rice, T.R. (JRB)	70	X	X	X	X	X												X									
Rugh, R. (HC)	55	X			X				X	X									X			X					
Scarborough, B.B. (RGA)	62	X			X		X									X											
Schroeder, J.H.	69	X			X										X												
Schroeder, J.H.	69	X			X										X												
Schroeder, J.H.	69	X			X										X												
Schroeder, J.H. (SH)	72	X			X										X												
Schroeder, J.H.	79	X													X												
Schroeder, J.H.	80	X			X										X	X											

Appendix II. Reference summary table

		General sorts					Effect endpoints												Modifying factors								
Author	Year	Fish	Invertebrates	Other	Acute	Chronic	Mortality	Pathophysiology	Reproduction	Development	Growth	Life span	Metabolism	Population	Genotoxicity	Behavioral	Cell culture	Other	Dose rate	RBE	Life stage	Phylogeny	Temperature	Salinity	Oxygen	Cell cycling	DNA repair
Setlow, R.B.	82			X																							X
Shechmeister, I.L. (+)	62	X			X		X	X																			
Slobodkin, L.B. (SR)	56		X											X				X									
Squire, R.D.	73		X		X										X												
Squire, R.D.	70		X		X		X		X			X															
Strand, J.A. (+)	73	X				X		X													X						
Strand, J.A. (+)	73	X				X	X			X	X																
Styron, C.E.	71		X		X		X		X											X	X						
Suyama, I. (+)	80	X				X									X												
Suzuki, J. (NE)	83		X		X		X				X								X			X					
Templeton, W.L.	66	X				X	X			X											X						
Templeton, W.L. (+)	71																	X	X								
Templeton, W.L.	76	X	X															X									
Templeton, W.L.	80	X	X			X								X	X			X									
Till, J.E. (+)	76	X				X	X			X									X								
Trabalka, J.R. (CPA)	77	X				X			X	X					X												
Tsytsugina, V.G.	73	X				X									X												
Tsytsugina, V.G.	79														X		X										
Ulrickson, G.U.	73	X			X			X																			
Upton, A.C. (+)	60			X	X		X	X				X															
Wadley, G.W. (ADW)	71	X			X		X				X										X		X				
Walden, S.J.	73	X				X				X																	
Ward, E. (+)	71	X			X		X														X						
Watson, D.G. (WLT)	73	X	X			X												X	X								
Welander, A.D. (+)	48	X			X		X	X	X	X	X										X						
Welander, A.D.	54	X			X		X														X						
Whicker, F.W.																		X									
White, J.C. (+)	67	X	X				X				X										X		X	X			
Willis, D.L.	80			X	X		X																X				
Willis, D.L. (WLL)	76			X	X		X	X																			

Appendix II. Reference summary table

		General sorts					Effect endpoints												Modifying factors								
Author	Year	Fish	Invertebrates	Other	Acute	Chronic	Mortality	Pathophysiology	Reproduction	Development	Growth	Life span	Metabolism	Population	Genotoxicity	Behavioral	Cell culture	Other	Dose rate	RBE	Life stage	Phylogeny	Temperature	Salinity	Oxygen	Cell cycling	DNA repair
Woodhead, D.S.	70	X				X				X								X	X								
Woodhead, D.S.	73	X				X												X	X								
Woodhead, D.S.	73	X	X			X												X	X								
Woodhead, D.S.	76	X			X										X		X										
Woodhead, D.S.	77	X				X			X	X					X				X								
Woodhead, A.D. (+)	79	X			X			X									X	X									
Woodhead, D.S.	79			X														X	X								